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Optimisation de l'extraction des composés phénoliques à partir des noyaux du fruit de la pêche en vue de leur valorisation

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Soutenu le : **15 Septembre 2022**

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End of Cycle Memory
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Theme

Optimization of the extraction of phenolic compounds from the kernels of the fruit of the peach in view of their valorization

Presented by :
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DEDICATION

This study is wholeheartedly dedicated to our beloved parents, who have been our source of inspiration and gave us strength when we thought of giving up, who continually provide their moral, spiritual, emotional, and financial support. To our brothers, sisters, relatives, mentor, friends, and classmates who shared their words of advice and encouragement to finish this study. And lastly, we dedicated this book to the Almighty God, thank you for the guidance, strength, power of mind, protection and skills and for giving us a healthy life. All of these, we offer to you.

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List of abreviations

Abs: Absorbance

TPC: Total Phenolic Compounds

DPPH: 1,1-diphenyl-2-picryl-hydrazyl

ABTS: 2.2¹ -Azinobis (3 - ethylbenzothiazoline - 6 - sulfonic acid) diammonium salt

GAE: Gallic acid equivalent

AAE: Ascorbic acid equivalent

EQ: Quercetin equivalent

DM: Dry Matter

UAE: Ultrasound Assisted Extraction

ROS: Reactive Oxygen Species

GPX: Glutathione peroxidase

GSH: Glutathione

SOD: Superoxide dismutase

PH: Hydrogen potential

TCA: Trichloroacetic acid

PR: Reducing power

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Introduction

1 Introduction:

Phenolic compounds are common dietary phytochemicals found in fruits, vegetables and grains. Epidemiological evidences have suggested that food phenolics may have protective effects against degenerative diseases (**Mazza, 2000**). Most of the beneficial characteristics of phenolic compounds have been ascribed to their antioxidant activity which is a fundamental property important to life (Rice-Evans et al., 1997).

Peach (*Prunus persica* L.), one of the traditional fruits in China, is a popular fruit mainly due to its nutrient–rich, beautiful appearance, delicate flesh, juicy, and aromatic flavor **(Chang *et al.*, 2000).***P. persica* has become a product of interest in recent years because of its nutritional and health benefits. Effectively, several studies have shown that these fruits are considered to be a rich source of antioxidants which exhibited anti-carcinogenic, anti-bacterial, anti-viral, anti-tumor and anti-inflammatory properties which overcome some of the degenerative diseases that affect humans. In addition, peach have been used in the folk medicine to treat hemorrhages, infertility, eye inflammation, and spasm **(Doymaz, 2004;Jiménez *et al.*, 2008;Hussain *et al.*, 2011;Sharma *et al.*, 2014).**

For the past two decades, there has been a revival of the use of renewable resources. Due to the limited reserves of fossil fuels and the broad availability of renewable resources, these are especially appealing as a source of materials and energy. The integral utilization of crops and the use of waste streams in certain industries will grow in importance leading to a more sustainable way of producing materials. Peach industry produces a huge amount of waste, mainly represented by the seeds. Peach seeds, more precisely the kernels are reported to be a rich source in bioactive compounds such as polyphenols, which show a prominent antioxidant activity **(Adil *et al.*, 2007;Redondo *et al.*, 2017;Redondo *et al.*, 2018).** From all the valuable biomass extractives, polyphenols are a widespread group of secondary metabolites found in all plants, representing the most desirable phytochemicals due to their potential to be used as additives in food industry, cosmetics, medicine, and others fields. At present, there is an increased interest to recover them from plant of spontaneous flora, cultivated plant, and wastes resulted in agricultural and food industry. **(Talmaciu *et al.*, 2015)**

Recently, interest in novel and green techniques like Microwave-assisted extraction, supercritical fluid extraction, and ultrasound-assisted extraction to valorize by-products has increased, as they comply with standards set by Environmental Protection Agency, USA. These include less hazardous chemical synthesis; designing safer chemicals, safe solvents

auxiliaries, design for energy efficiency, use of renewable feedstock, reduce derivatives, design to prevent degradation **(Azmir *et al.*, 2013;Rombaut *et al.*, 2014).** Then the ultrasound-assisted extraction of bioactive compounds has been proposed for the valorization of peach kernels**.** Among these, ultrasound-assisted extraction (UAE) is an inexpensive, because of low instrumental requirements, simple, and efficient alternative to conventional extraction techniques. The enhancement in extraction obtained by using ultrasound is mainly attributed to the effect of acoustic cavitations produced in the solvent by the passage of an ultrasound wave (**Wang *et al.*, 2008**).

Thus, the aim of this study is to use the ultrasound-assisted extraction (UAE) for the optimization of phenolic compounds recovery from peach kernels using single factor experiment as experimental design. Five extractions parameters namely effect of solvent (60 acetone, 60% ethanol and 60% methanol), effect of acetone concentration (40%, 60% and 80%; v/v), extraction time (3, 5, 10, 15, 30 and 60 min), extraction temperature (25, 35, 45, 50 and 60°C) and solid/liquid ratio (50, 100, 250, 500 and 750 mg) were tested on the extraction of total phenolic compounds and total flavonoid compounds and on the antioxidant activities (DPPH-radical scavenging activity; DPPH-RSA, ABTS-radical scavenging activity; ABTS-RSA, reducing power; RP and total antioxidant activity; TAA). To the best of our knowledge, this is the first report on the optimization of the extraction of phenolics from peach kernels.
Our manuscript is subdivided in several parts, the first one is the bibliographic part comporting somme generalities on peach kernels and oxidative stress, the second part in which was discussed the material and methods used and the different results obtained and finally a conclusion and perspectives.

Bibliographic synthesis

Chapter I Generalities on peach plant (Prunus persica)

2 Chapter I: Generalities on peach plant (*Prunus persica* L.)

2.1 Description of the peach plant (*Prunus persica* L.)

Peach (*Prunus Persica* L.) is a plant cultivated in the warm temperate regions of the northern and southern hemispheres, belonging to the Rosaceae family. It is a deciduous tree up to 10 m high with greyish or ash colored bark. The peach fruit develops from a single ovary that matures both into a fleshy, juicy exterior that forms the edible part of the fruit and a hard interior, called the pit **(Kant *et al,* 2018)**. Peaches are very broad in color (skin color; red, purple-red, pink, orange and variegated), (fruit color; red, yellow, white or red-yellow, red-white), texture, shape (full round, slightly flattened or fully flattened) **(Yılmaz *et al*, 2021)**

Figure 1:Photographed picture of Prunus Persica plant(Binette and Jardin, le monde)

2.2 Taxonomic classification of peach

The Taxonomic classification of peach **(Kant *et al*., 2018)** is the following:

Kingdom: Plantae

Sub-kingdom: Tracheobionta

Super division: Spermatophytes

Department: Magnoliophyta

Class: Magnoliopsid

Subclass: Rosidae

Order: Rosales

Family: Rosaceae

Subfamily: Amygyloideae (Prunoids)

Gender: Prunus

Species: *Prunus persica* L.

2.3 History and origin

About 3,000 years ago, peach was moved from China to all temperate and subtropical climates of the Asian continent, and about 1,500 to 2,000 years ago, to Japan. From Asia, it spread to Persia (now Iran) and from there to all of Europe over 2,000 years ago. This fruit was introduced to America by the Spaniards and Portuguese during the 16th century, from the tropical highlands of South and Central America, to the humid subtropics of Florida and southern Brazil and to the most northern United States and southern Canada. Then, it was quickly adopted by the Indians and spread across a wide spectrum of the region. There were probably several introductions from different parts of Spain, the Canary Islands, Portugal and even the South Pacific, as some genotypes have adapted well to humid subtropical areas. Propagation by seed was the main source of plants until the first half of the 19th century in the United States and Europe and until the middle of the last century in Central and South America. Thus, there are many varieties of peaches that have undergone centuries of selection for adaptation across Europe, America, Asia and Japan **(Byrne *et al*., 2012)**

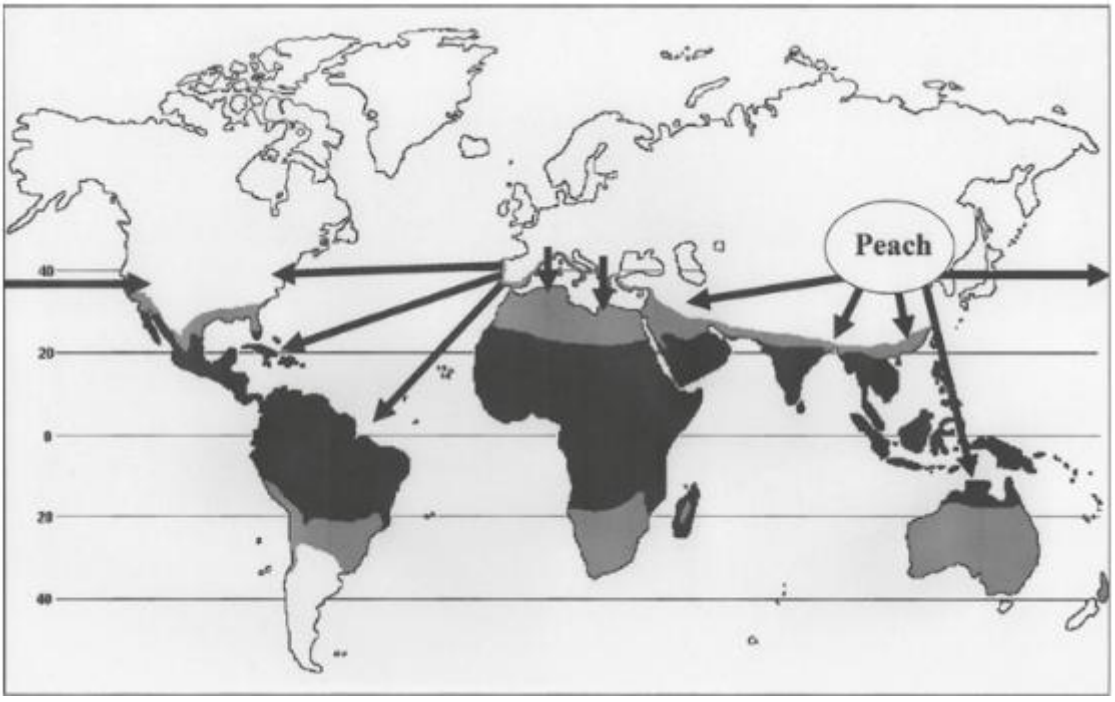


Figure 2:Distribution of peach throughout the world(Byrne *et al*., 2012)

2.4 World production

Production of peach fruit is estimated at 22 million tons each year. China tops the list of world's leading peach producers with 12.0 million tons, followed by Italy, Spain and United States with 1.64, 1.34 and 1.19 million tons respectively. Europe, Italy, Spain, Greece and France together produce 42% of world production. According to FAO statistics (2011), the largest producer of peaches and nectarines is Italy with 1,474,337 tonnes, followed by Spain (1,129,300 tonnes), Greece (810,000 tonnes) and France (313 300 tonnes). Spain is the major exporter of this fruit while Greece is the main processor of peaches in the EU **(Habib, 2015).**

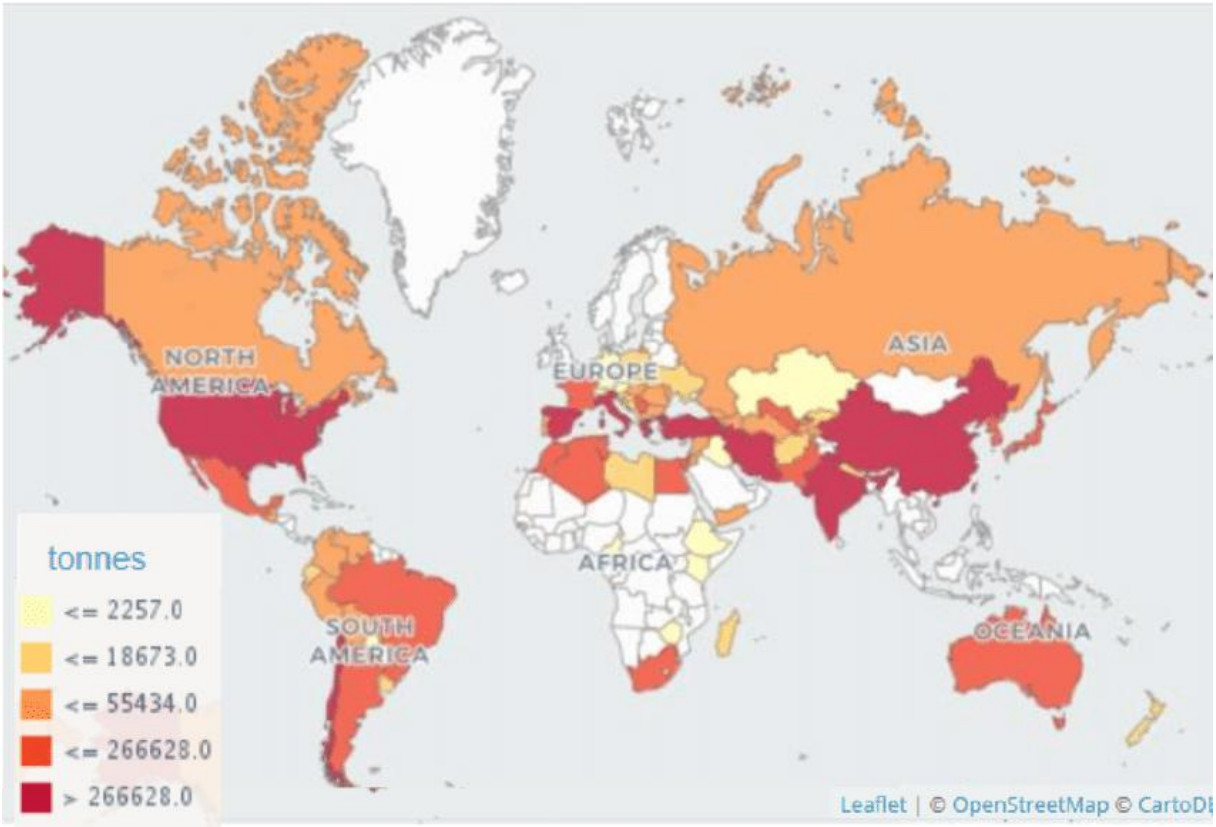


Figure 3:Global map of peach and nectarine production.(Obi*et al.*,2018)

Tableau I:World peach production(Faostat Database (FAO) United Nations.,2020)

Rank	Country	Production (million tons)
1	China	12
2	Italy	1.64
3	Spain	1.34
4	USA	1.19
5	Greece	0.70
6	Turkey	0.61
7	Iran	0.52
8	Chile	0.34
9	France	0.30
10	Argentina	0.28

2.5 Local production

In Algeria, peach cultivation can succeed anywhere in the coastal zone where frost is limited. Low altitude areas (300m) such as in the Sahel and coastal lands, may be suitable. At this altitude it is generally advisable to use late flowering cultivars to avoid frost damage. The cultivated area was approximately 10,146 ha and the production was approximately 186, 44 tons in 2020, an overall productivity of 3 to 4 t/ha depending on the different cultivated areas, this production still remains low compared to the potential of our orchards. Regarding the main peach cultivars in Algeria, two different groups are distinguished according to the color of the fruit pulp, cultivars with white pulp and cultivars with yellow pulp **(Belhadj, 2003; FAO : 2020)**

Tableau II:Spatial distribution of the cultivated area and the production of peach fruit in Algeria(Belhadj, 2003)

Region	East	Center	West	Total
Total area (ha)	2181	4094	3870	10146
Total production (t)	5835.8	12817.2	4288.5	22941.5
Productivity (t/ha)	3.8	4	1.7	3.2

2.6 Peach industry

The major change experienced on the technology of peach production in the last two decades has been the high number of cultivars from different breeding programs introduced a commercial scale by the growers. Innovation through development and commercialization of new and improved cultivars has become the key strategy to enhance economic sustainability and profitability of peach industry **(Iglesias, 2013).**

Every year, millions of thousands of peach fruit (*Prunus persica* L.), around 40% to 45% of world production are used to produce juices, nectar, jam, canned peaches, sauce, baby food, wine, and vinegar. Consequently, thousands tones of fruit waste are discarded, Among these wastes, we mainly distinguish the kernels which are rich in phytochemicals like phenolics and carotenoids that have high economic added value, due to their remarkable biological activities that find many applications in food, cosmetic and pharmaceutical industries **(Madrau *et al*., 2009;Chavan *et al* , 2013).**

2.7 Biochemical composition of the peach kernels

The peach kernels have an interesting nutritional value. They are rich in lipids, and are used as a source of excellent quality oil which has wide applications in the cosmetics industry. To valorize and effectively use these agro-food residues, an in-depth knowledge of their properties and their biochemical composition is essential. The presence of cyanide in the peach kernels corresponds to most of the published information relating to the composition of the grain. The cyanide glycoside in the peach kernel, also called amygdalin, can be around 710-720 mg/kg **(Haque *et al*, 2002; Pelentir*et al*, 2011).**

Peach kernels have a low saturated fatty acid content, however, they have significant amounts of oleic and linoleic acids, about 55 and 77%, respectively **(CALGAROTO *et al*.,2005).** Firestone (2006) obtained values between 61 and 70% for oleic acid and between 15 and 29% for linoleic acid**(Firestone, 2006)**

Tableau III:Fatty acid composition in peach kernel oil(Pelentir *et al*., 2011).

Fatty acid	Content in g /100g
C14:0 Miristic	/
C16:0 Palmitic	8.39
C16:1 Palmitoleic	0.32
C18:0 Esteric	1.18
C18:1 trans Elaidic	/
C18:1 Oleic	41.1
C18:2 trans linoelaidic	/
C18:2 Linoleic	48.4
C18:3 trans linolenic	/
C18:3 Linolenic	0.3
C20:0 Araquidic	0.2
C20:1 Gadoleic	0.1
C22:0 Behenic	0.1

The protein content and moisture of peach kernels reported by Salem and al. (1974) were 2.677% and 3.1% respectively **(Salem *et al.*, 1974).**

The fiber content of peach kernels was estimated at 1.86% according toShahid and Dildar**(Shahid *et al.*, 2011).**

Tableau IV:Composition of different minerals in peach kernels(Shahid *et al*, 2011)

Mineral	Content in mg/100g
Sodium	16
Potassium	50
Zinc	4.3
Copper	0.06
Iron	0.30

2.8 Therapeutic properties of Peach plant

2.8.1 Beneficial effects on health

Peach has been part of the human diet for hundreds of years, it has an incredible richness in phyto-chemical compounds such as phenolic compounds, carotenoids, vitamins, volatile compounds and organic acids. These compounds are known for their antimicrobial, anti-inflammatory and antioxidant properties, exerting a number of beneficial effects on cells through the scavenging of free radicals and by participating in cell signaling pathways. Irrefutable, these compounds have demonstrated many preventive effects on a wide range of age-related chronic diseases such as hypertension, obesity, cardiovascular, neuro-degenerative and oncological diseases **(Bento *et al* ., 2020).**

Some studies have shown that the phenolic compounds present in this fruit, including anthocyanins, anthocyanidins and catechins which has an inhibitory effect on chemical carcinogenesis. They also induce apoptosis of cancerous cells of the prostate**(Kampa *et al.*, 2000;Chung *et al.*, 2001)**, of the breast, and stop the proliferation of endothelial cells. **(Byrne *et al.*, 2007)**

2.8.2 Use of peach in traditional medicine

Peach (*Prunus persica* L.) is a Chinese herbal medicine, which was first mentioned in the Qing Dynasty medical manual, Golden Mirror of Medicine, its pits were traditionally used to promote blood circulation and eliminate blood stasis **(Han *et al.*, 2021)**

In traditional Chinese folk medicine, the flowers of *Prunus persica* L. have been used as a purgative or diuretic, and have also been used in cosmetology **(Han *et al*., 2015)**

Traditionally, the peach plant was implemented mainly for the treatment of whooping cough and bronchitis, the leaves were used to treat digestive ailments, mainly prepared in the form of tea, the oil extracted from the seeds helped to strengthen hair growth. The most common use of peach in traditional medicine was cooked peaches which were effective in relieving stomach pain, treating ulcers, intestinal inflammation and colitis **(Hussain *et al*, 2015)**

ChapterII

Generalities on polyphenols and oxidative stress

3 Chapter II: Generalities on polyphenols and oxidative stress:

3.1 Definition of polyphenols

Polyphenols are natural compounds synthesized exclusively by plants, with chemical characteristics related to phenolic substances and strong antioxidant properties **(Singla *et al*., 2019)**. Polyphenols are compounds that have more than one phenolic hydroxyl group attached to one or more benzene rings. The term is somewhat misleading since it tends to make people think of polymers of individual phenol molecules. Of course such polymers exist, phenolic compounds are characteristic of plants and as a group they are usually found as esters or glycosides rather than as free compounds. It is important to realize this for the extractions of phenols from plants tissues **(Vermerris *et al*., 2007).**

3.2 Biosynthesis of polyphenols

Understanding polyphenol biosynthetic pathways can help design foods which are rich in polyphenols with health benefits.Like all phenolic compounds, phenolic acids such as gallic acid and cinnamic acid are considered to be metabolites of the shikimate pathway. Biosynthesis of complex polyphenols such as flavonoids is linked to primary metabolism through plastid and mitochondrial derived intermediates, each requiring export to the cytoplasm where they are incorporated into separate parts of the molecule. The aromatic ring B and the chromane ring are considered to originate from the amino acid phenylalanine, itself a product of the shikimate pathway, whereas Ring A from three units of malonyl-CoA. These three malonyl-CoA units are added through sequential decarboxylation and condensation reactions which initiates flavonoids biosynthesis **(Tsao, 2010)**

3.3 Classification of polyphenols

To date, about 8000 types of polyphenols have been identified and reported in the scientific literature. Polyphenols are subcategorized into phenolic acids (hydroxybenzoic and hydroxycinnamic acids), flavonoids (flavones, flavonols, isoflavones, flavanones, anthocyanins), stilbenes (resveratrol, piceatannol), lignans (sesamol, pinoresinol, sinol,

enterodiol), and others including tannins (hydrolyzable, non-hydrolyzable and condensed tannins), lignins, xanthones, chromones, anthraquinones. Dietary polyphenols represent a wide range of secondary metabolites, mainly derived from phenolic acid, catechins, flavones and isoflavones **(Prabhu*et al*., 2021)**

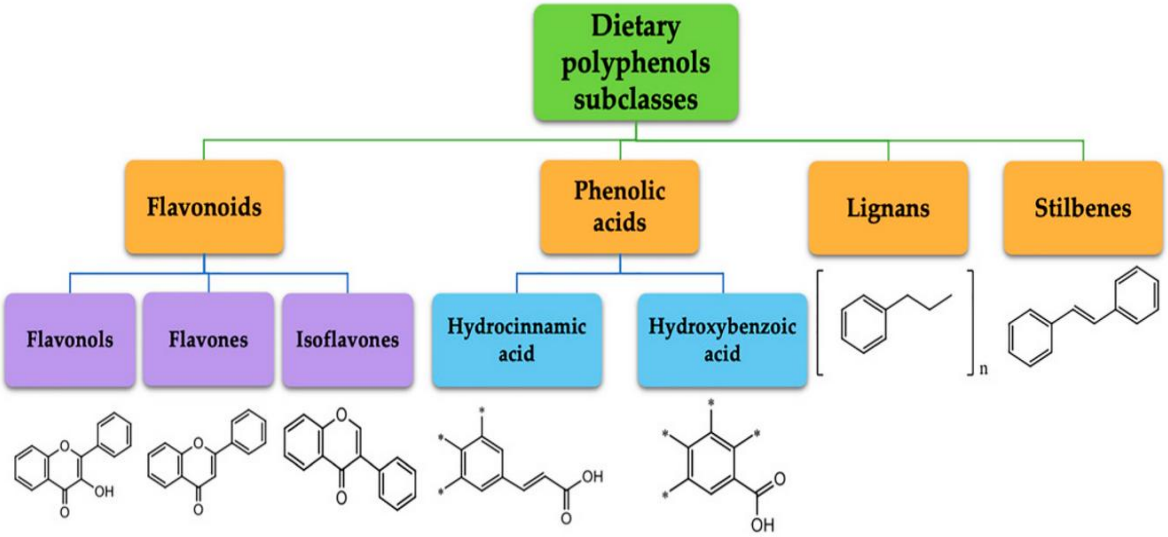
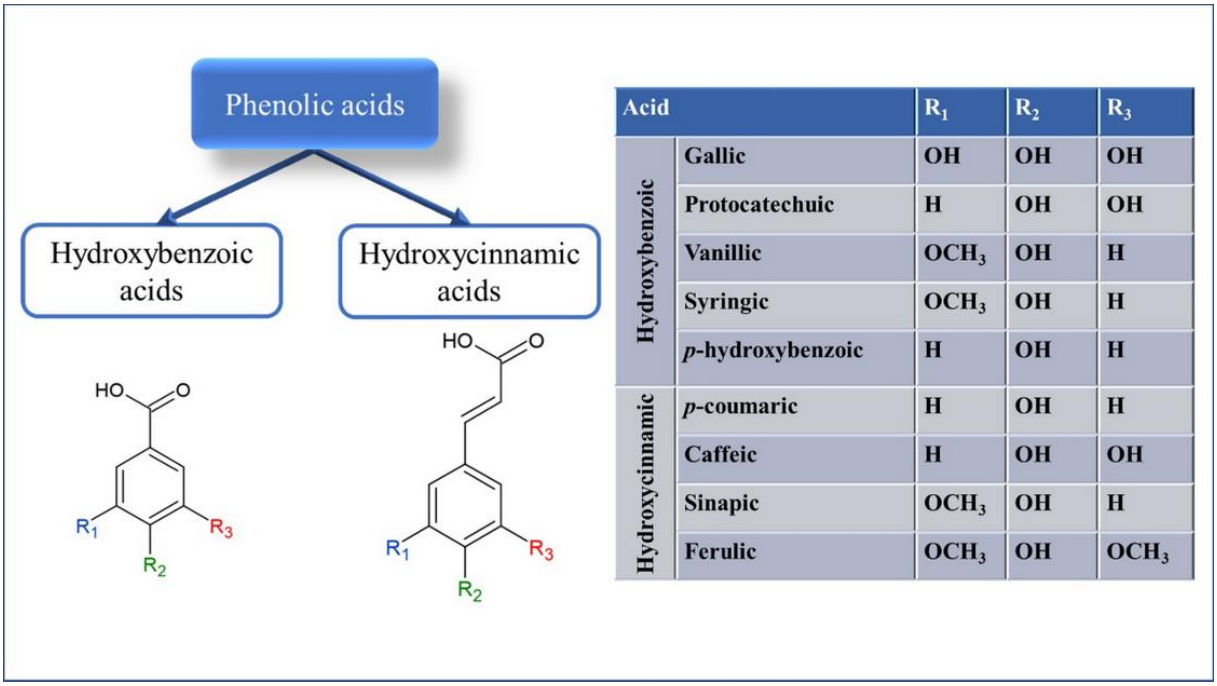


Figure 4:General classification of polyphenols

(Truzzi *et al*, 2021)

3.3.1 Phenolic acids

Phenolic acids are composed of aromatic rings with a carboxylic acid group (-COOH). These phenolic compounds represent the main class of phenolic compounds of plant. Phenolic acids are subdivided into hydroxybenzoic and hydroxycinnamic acids. Hydroxybenzoic acid is composed of C6-C1, a derivative of benzoic acid (C7H6O2). Salicylic acid, vanillic acid, protocatechin acid, gallic acid, benzoic acid, and ellagic acid are categorized under the subcategory of hydroxybenzoics. Hydroxycinnamic acid represents the class of aromatic acids (C6-C3), derived from cinnamic acid. Caffeic acid, ferulic acid, coumaric acid, sinapinic acid and and cinnamic acid are common examples of hydroxycinnamic acid **(Singla *et al*., 2019)**



	Acid	R_1	R_2	R_3
Hydroxybenzoic	Gallic	OH	OH	OH
	Protocatechuic	H	OH	OH
	Vanillic	OCH_3	OH	H
	Syringic	OCH_3	OH	H
	p-hydroxybenzoic	H	OH	H
Hydroxycinnamic	*p*-coumaric	H	OH	H
	Caffeic	H	OH	OH
	Sinapic	OCH_3	OH	H
	Ferulic	OCH_3	OH	OCH_3

Figure 5:Main phenolic acid compounds

(Paucar-Menacho *et al*., 2022)

3.3.2 Flavonoids

A flavonoid is a main group of plant metabolites of polyphenolic compounds. From a structural point of view they are composed of 15 carbon atoms, they include 2 aromatic rings linked by a chain with 3 carbon atoms **(Harborne *et al*, 2013)**

Flavonoids can be divided into several categories depending on the C ring to which the B ring is attached. The different categories of flavonoids including flavones (chrysin, apigenin, baicalein), flavonols (quercetin, kaempferol), isoflavones (daidzein , glycitein), flavan-3-ols (gallocatechin, catechin, epicatechin), flavanones (hesperetin, naringenin) etanthocyanidins (delphinidin, peonidin, cyanidin, pelargonidin) are the major class of flavonoids **(Rasouli *et al*, 2019).**

3.3.2.1 Flavonols

Flavonols are a category of the flavonoid family, composed of double bond between C2-C3 and carbonyl C4. Flavonols include quercetin, kaempferol and myricetin. New evidence

from clinical trials has shown that flavonols have the potential to pre-treat and prevent cardiovascular disease, regenerate human gum cells and other heart diseases **(Singla *et al*., 2019).**

3.3.2.2 Isoflavones

Isoflavones are a type of non-steroidal phyto-estrogens, these compounds derived from plants, they are obtained through the phenyl-propanoid pathway which helps to produce flavone groups in higher plants **(Nikolić *et al*., 2017)** Researches suggests potential for using isoflavones to treat menopause-related symptoms as well as being thought to have chemo-protective properties **(Rasouli *et al*., 2019).**

3.3.2.3 Flavanones

The flavanones structure is characterized by a benzopyranone core substituted at the C2 position with possible substitution on the aryl backbone of the benzopyranone core **(Nibbs*et al*., 2012)**.Citrus fruits, certain aromatic plants and tomatoes contain quantities of flavanones which form a small part of flavonoids. They are considered as an important element of human health apart from their flavoring properties. Eriodyctiol from lemons, hesperidin from orange and naringenin from grapefruit constitute the flavanones **(Calderón-Oliver *et al*., 2018).**

3.3.2.4 Anthocyanidins

Anthocyanidins are the pigments mainly responsible for the color (red, pink, purple) of fruits and vegetables **(Chaovanalikit *et al*., 2004)**. The different colors of fruits, vegetables and flowers are due to the presence of anthocyanidins in their epidermis. Different fruits and vegetables including beets, berries, strawberries and cherries are a good source of anthocyanidins. It has been concluded that the consumption of strawberries will help reduce thrombotic inflammatory responses **(Welch *et al*., 2008).**

3.3.3 Other classes of polphenols:

Other polyphenols including stilbenes (resveratrol, piceatannol), Lignans (sesamol, pinoresinol, sinol, enterodiol), and others including tannins (hydrolysable, non-hydrolysable and condensed tannins), lignins, have a wide range of therapeutic and industrial applications according to their nature of action **(Prabhu *et al*., 2021).**

3.3.3.1 Lignin:

In many plant tissues there is a group of complex organic compounds, these compounds are called lignin. Found in plants and trees, lignin is particularly important as it helps in cell membrane formation **(Malutan *et al*., 2008)**. They are important because they help provide strength and rigidity to trees. Lignin is a heterogeneous polymer which is obtained from few signal precursors which are cross-linked in different shapes **(Singla *et al*., 2019)**. There are three types of crosslinks, which are obtained from phenylpropane, these crosslinks are coniferyl alcohol, sinapyl alcohol and paracoumaryl alcohol **(Malutan *et al*., 2008).**

3.3.3.2 Silymarin:

Silymarin is a type of the flavonolignans which has antioxidant properties. This type of lignin is obtained from milk thistle seeds which are obtained from certain varieties of daisies and other herbaceous plants **(Radko *et al*., 2007)**. For ages, this lignin has been used to treat ailments related to the gallbladder and the liver. Some researchers also suggest its use against cirrhosis and jaundice. Some also claim that it is also used to treat diabetes specifically type 2 and it is also used to lower cholesterol levels **(Křen *et al*., 2005).**

3.3.3.3 Stilbenes

They are organic compounds that have a compact form with a central ethylene moiety and a phenyl group. The phenyl group is located at the ends of the carbon double bonds ***(*Chou *et al*., 2018).** Their biological activity and overall health benefits are of major interest for several studies **(Shen *et al*., 2009).**

3.4 Biological activities of polyphenols

3.4.1 Antioxidant activity

Among the notable biological activities of phenolic compounds, antioxidant activities have been widely studied, including scavenging of free radicals, inhibition of lipid oxidation, reduction of hydroperoxide formation, and so on **(Sato *et al*., 1996)**. Numerous in vitro experiments have proven that phenolic compounds are generally the main contributors to the antioxidant capacities of plants. Among these contributors, we mainly distinguish rosmarinic, ferulic, caffeic, chlorogenic, vanillic acid, p-hydroxybenzoic acid, p-coumaric acid, protocatechuic acid, etc. Polyphenols may also function as antioxidants through their effects on plasma, membranes, transcription factors and in vivo enzymatic activities **(Ślusarcz *et al*., 2009;Roy *et al*., 2010).**

The mechanism of the antioxidant activity of polyphenols **(Kurek-Górecka *et al*., 2013)**consists of :

- Inhibit the activity of enzymes and thus inhibit the appearance of reactive oxygen species (ROS).
- Metal ion chelators involved in the process of creating free radicals.
- Scavenge reactive oxygen species (ROS), thereby interrupting the cascade of reactions leading to lipid peroxidation.
- Synergistic action with other antioxidants

3.4.2 Anti-inflammatory activity

Several polyphenols have been shown to reduce the incidence of a wide variety of inflammatory diseases in vitro and in vivo. Although the mechanism of action is strictly dependent on the class of polyphenols considered, several phytochemicals act as anti-inflammatory agents through the alteration of the nuclear factor κB (NF-κB) pathway, which plays a central role in the development of the inflammatory response by promoting the expression of inflammatory disease and the expression of adhesion molecules, cytokines and other pro-inflammatory mediators. This factor is present in an inactive form in the cytoplasm of cells and, following stimulation by pathogens (bacteria), cytokines or pro-oxidants such as ROS, NF-κB, it dissociates from its inhibitory protein IκBα and moves into the nucleus, where it modulates the transcription of various cytokines. Transcription of a variety of cytokines and pro-inflammatory molecules and inhibition of the NF-κB pathway as well as its nuclear translocation, are considered as a key points to reduce cell damage due to the chronic

inflammation that occurs in pathological conditions. Several studies have confirmed that NF-κB is one of the preferential molecular targets of a multitude of polyphenols ***(*Sangiovanni *et al*., 2020).**

3.4.3 Antimicrobial activity

The main antimicrobial power of polyphenols lies in the fact that they can pass through the gastro-intestinal system without being absorbed, thus affecting the intestinal microbiota. This can have two consequences: first, the polyphenols are modified in their active form; second, they change the composition of the gut microbiota, likely by inhibiting pathogenic bacteria and enriching beneficial bacteria. Thus, polyphenols have a significant impact on human health **(Abbas*et al*., 2017)**.

Among the main mechanisms responsible for the antimicrobial activity of polyphenols, we distinguish in particular the inhibition of enzymes by oxidized compounds, possibly through reactions with proteins through SH groups or through non-specific interactions **(Mason, 1987).**

3.5 Free radicals

3.5.1 Definition

Free radicals can be defined as molecules or molecular fragments containing one or more unpaired electrons in atomic or molecular orbitals. These unpaired electron(s) generally impart a considerable degree of reactivity to the free radical **(Pala *et al*., 2008).** Oxygen-derived radicals represent the most important class of radical species generated in living systems. The harmful effect of free radicals causing potential biological damage is called oxidative stress or nitrosative stress **(Valko *et al*., 2007).**

3.5.2 Sources of free radicals

3.5.2.1 Exogenous sources

They come from pollution, tobacco, ozone, heavy metals, food pollutants (fertilizers, additives), saturated fats from food, excess sugars, alcohol, drugs, prolonged exposure to the sun and ultraviolet rays **(Favier, 2003).**

3.5.2.2 Endogenous sources

Free radicals are most often formed from oxygen, hence their name: reactive oxygen species (ROS). The endogenous origin of ROS is mainly the mitochondrial respiratory chains of the cells of aerobic organisms (about 2% of the oxygen consumed at the mitochondrial level is transformed into particularly reactive ROS), dysfunction of the enzymatic system or lack of antioxidants in the organism and the inflammatory reaction which is an important source of oxygenated radicals produced directly by the activated phagocytic cells, which are the seat of a phenomenon called "Oxidative explosion" consisting of the activation of the NADPH oxidase complex **(Puppo *et al.*, 1988).**

3.6 Oxidative stress

3.6.1 Definition

Oxidative stress is defined by an imbalance between the production of radical (or reactive) oxygen species (ROS) and the cellular antioxidant capacities. ROS have long been considered toxic by-products of normal oxygen metabolism and implicated in many pathologies **(Migdal *et al.*, 2011).**

3.6.2 Consequences of oxidative stress:

The consequences of oxidative stress will be extremely variable depending on the dose and cell type. Light stresses will increase cell proliferation and the expression of adhesion proteins, medium stresses will facilitate apoptosis, while strong stresses will cause necrosis and violent stresses will disorganize the membrane leading to immediate lysis. Other biological disturbances are observed following oxidative stress: decrease in membrane fluidity, receptor abnormalities, decrease in insulin sensitivity, disturbance of cellular immunity, fibrosis, lipid deposits, weakening muscle, even neuronal death or appearance of mutations. Many pathological abnormalities are also induced by oxidative stress: mutations, carcinogenesis, fetal malformations, abnormal protein deposits, fibrosis, formation of auto-antibodies, oxidized lipid deposits, immunosuppression…etc. **(Favier, 2006).**

3.6.3 Systems of defense against oxidative stress

3.6.3.1 Enzymatic antioxidants system

There are several antioxidant enzymes **(Bensakhria, 2018)**

- **Superoxide dismutase (SOD):** ubiquitous metalloenzyme, it eliminates the superoxide anion by disproportionation.
- **Catalase**: ubiquitous heme enzyme located inside red blood cells, it eliminates H2O2 by disproportionation.
- **Glutathione peroxidase / Glutathione reductase (GPx/GR) system:** ubiquitous selenoprotein (7 isoforms) eliminates 70% of organic proxies and 94% of H2O2 by reduction.
- **Thioridoxine peroxidases (Trx)**: NADPH-dependent selenoenzymes eliminate H2O2, ROOH, ONOO–) by reduction.

3.6.3.2 Non-enzymatic antioxidants system

Non-enzymatic antioxidants include **(Ahmad *et al*., 2010):**

- **Vitamin E:** in the form of α-tocopherol (the most active and the most absorbed), a major antioxidant in lipid structures, it also has another action, the neutralization of O2.
- **Vitamin C:** ascorbic acid, it is a reducing and chelating agent in the form of dehydro-L-ascorbic acid (DHA), it reacts directly with free radicals and eliminates H2O2.
- **Provitamin A (carotenoids)**: β-carotene: precursor of vitamin A, it interrupts the process of lipid peroxidation.
- **Other vitamins**: vitamin P (flavonoids), Coenzyme Q10.
- **Trace elements**: Se, Zn as cofactors of GPx, SOD1, SOD3 respectively. Transport proteins by sequestration of metals involved in the generation of ROS, for example: transferrin and iron.
- **Glutathione**: is a cofactor of the enzyme GPx. It is a natural tripeptide, water-soluble L-γ-glutamyl-Lcysteinylglycine (cytoplasm, nucleus, mitochondria), whose GSH constitutes 90% of its total content. Glutathione is the cofactor of many antioxidant enzymes (GPx) it allows the reduction of oxidized proteins by conjugation to electrophilic species.

3.6.3.3 Other mechanism

When the antioxidants prove to be "insufficient" or defective, the cell has another means of repairing the damage caused and thus preventing the risk of mutagenesis, possibly of cancer. DNA repair involves several mechanisms **(Bensakhria, 2018)**, for example:

- Mismatch repair system.
- Repair by excision or resynthesis.
- Nucleotide excision repair.
- Recombination.

Materials and methods

1 Chemicals and reagents

Acetone, ethanol and methanol were obtained from Honeywell (Seelze , Germany) . Trolox (6 - hydroxy - 2.5.7.8 - tetramethyl chromane - 2 - carboxylic acid) , ABTS (2.2¹ - Azinobis (3 - ethylbenzothiazoline - 6 - sulfonic acid) diammonium salt) were purchased from Sigma-Aldrich (Fisher scientific , Fair Lawn , NJ , USA) . DPPH (2,2 - diphenyl - 1-picrylhydrazyl) . disodium hydrogen phosphate (Na_2HPO_4) , sodium dihydrogen phosphate (NaH_2PO_4) , 4- hydroxy - 3 - methoxybenzaldehyde (vanillin) , ferric chlorid ($FeCl_3$) , gallic acid , catechin quercetin and trichloroacetic acid were purchased from Sigma - Aldrich Chemie GmbH (Steinheim , Germany) . Hydrochloric acid (HCl) , Sodium carbonate (Na_2CO3) were purchased from Prolabo (Loire , France) . Folin - Ciocalteu's phenol reagent , Potassium ferricyanide ($C_6N_6FeK_3$) and chloride aluminium ($AlC1_3$) were provided from Biochem-chemopharma (Loire , France) .

2 Plant material

The peach fruit samples used in this study were purchased from local market (Bejaia city) in spring 2022. The fruits were transferred to the laboratory, washed with water and dried with absorbent paper. In order to verify that peach fruits reached their maturity, we proceeded to the measure of the Brix index by a refractometer (ATAGO, Japan). The peach kernels were recovered from the stones of the peach fruits. The kernels were cut into small pieces and then lyophilized (Alpha1-4 LD plus, Christ, Osterode, Germany). The lyophilized kernels were ground in a blender to obtain fine pounder (figure 7). The powdered peach kernels were stored at 4°C until extraction.

Figure 6:Photographed picture of the powdered peach kernels.

3 Ultrasound-assisted extraction (UAE) of phenolic compounds from peach kernels

Phenolic compounds were extracted from powdered peach kernels using ultrasonic-assisted extraction (UAE) technique.

Ultrasound-assisted extraction was performed in an ultrasonic cleaning bath (2510E-DTH type, 42 kHz, 100 W, 41 Eagle Road, Danbury, CT 06813, USA) with a useful volume of 10 l (internal dimensions: 300x240x150 mm). Working frequency was fixed at 42 kHz.

Powdered kernels were placed into a volumetric flask (50 ml), mixed with extracting solvent (30 ml) and sonicated for different times at the required temperature. The temperature was controlled by circulating external water from a thermostatic water bath. After the extraction, the flask was removed from the bath and cooled to room temperature by cooling water. The peach kernel extracts were centrifuged at 5000 rpm for 10 min. The supernatants were filtered through filter paper. The filtered extracts were stored at 4°C until analysis. These extracts were used for the determination of the total phenolic compounds (TPC), total flavonoid compounds (TFC) and antioxidant activities (total antioxidant activity; TAA, DPPH radical-scavenging activity; DPPH-RSA, ABTS radical-scavenging activity; ABTS-RSA and ferric reducing power; FRP).

4 Experimental design

Single factor experiment was used to select the optimum conditions for extracting phenolic compounds from peach kernels. A total of six parameters were tested: extraction solvent (60% acetone, 60% ethanol and 60% methanol), acetone concentration (40%, 60% and 80%; v/v), extraction time (3, 5, 10, 15, 30 and 60 min), extraction temperature (25, 35, 45, 50 and 60°C) and solid/liquid ratio (50, 100, 250, 500 and 750 mg) were studied on which one parameter was varied at a time while the other parameters were fixed.

4.1 Selection of extraction solvent

Powdered peach kernels (750 mg) were extracted with 30 ml of 60% acetone, 60% ethanol and 60% methanol, respectively, in a volumetric flask (50 ml) and kept for sonication at 25°C for 30 min. After sonication, mixtures are centrifuged at 5000 rpm for 10 min. The supernatant were filtered through filter paper.

4.2 Effect of solvent concentration

Phenolic compounds were extracted from peach kernels (750 mg) using acetone (best extraction solvent determined previously) at different concentrations (40%, 60% and 80%; v/v), by sonication at 25°C for 30 min. The extracts were centrifuged and filtered.

4.3 Effect of extraction time

Peach kernels (750 mg) were macerated with 60% acetone (30 ml) and sonicated for different times (3, 5, 10, 15, 30 and 60 min) at 25°C. The extracts were centrifuged and filtered.

4.4 Effect of extraction temperature

Peach kernels (750 mg) were macerated with 60% acetone (30 ml) and sonicated for 30 min at different temperatures (25, 35, 45, 50 and 60°C). The extracts were centrifuged and filtered.

4.5 Effect of solid/liquid ratio

Different weightsofpeach kernels (50, 100, 250, 500 and 750 mg) were macerated with 60% acetone (30 ml) and sonicated for 30 min at 25°C. The extracts were centrifuged and filtered.

4.6 Determination of total phenolic compounds (TPC)

Total phenolic compounds (TPC) were determined with the Folin-Ciocalteu method. The Folin-Ciocalteu with yellow color is a mixture made up of phosphotungstic acid (H3PW12O40) and phosphomolybdic acid (H3PMO12O40). In a basic medium (sodium carbonate), the Folin-Ciocalteu reagent is reduced during the oxidation of phenolic compounds, to a mixture of blue oxides of tungsten and molybdenum **(Ribéreau-Gayon *et al*. 1982).**

The content of phenolic compounds was determined according to the protocol of **(Boizot & Charpentier, 2006)** with a slight modification. Peach kernel extract (200 µl) of with 1 ml of Folin-Ciocalteu's reagent (diluted 10 fold) and 800 µl of sodium carbonate Na2CO3 (7.5%). The mixture was incubated for 30 min at room temperature in the dark, and the absorbance was measured at 760 nm using UV-VIS spectrophotometer (Shimadzu UV mini1240, Suzhou Jiangsu, China). The intensity of the coloration is proportional to the quantity of phenolic acids present in the sample. The concentration of phenolic compounds was determined by referring to the calibration curve obtained under the same conditions using gallic acid (annex I) and results were expressed in mg gallic acid equivalents per 100 g of dry matter (mg GAE/100 g DM).

4.7 Determination of total flavonoids compounds (TFC)

The determination of total flavonoid compounds (TFC) was carried out by a colorimetric method according to the method of Kim et al **(Kim et al 2003)**. Extract (500 µl) was mixed with 150 µl sodium nitrite solution (5%) and 300 µl aluminum chloride (10%). After 5 min, 1 ml sodium hydroxide (1 M) was added. The absorbance of the mixtures was measured at 510 nm. The concentrations of flavonoids were determined by referring to a calibration curve performed with quercetin as standard (annex II) and results were expressed in mg quercetin equivalents per 100 g of dry matter (mg QE/100 g DM).

4.8 Antioxidant activities

4.8.1 DPPH-radical scavenging activity (DPPH-RSA) assay

1,1-diphenyl-2-picrylhydrazyl (*DPPH*$^{\bullet}$) is a stable free radical soluble in methanol. In this test the antioxidants reduce the *DPPH*$^{\bullet}$ having a violet color into a yellow compound; the intensity of the color is inversely proportional to the capacity of the antioxidants present in the medium to donate protons and electrons **(Kim *et al*., 2013).**

The DPPH-RSA was measured according to the method of Brand-Williams et al. **(Brand-Williams *et al*., 1995)**. Briefely, 900 µl of the DPPH solution (0.04 mg/ml in methanol) were added to 100 µl of the extracts. The mixtures were vortexed are then incubated in the dark at room temperature for 20 min. The decrease of absorbance was measured at 517 nm against a blank. The DPPH-RSA was determined by referring to the calibration curve made with Trolox and results were results were expressed in mg Trolox equivalents per 100 g of dry matter (mg QE/100 g DM).

4.8.2 ABTS-radical scavenging activity (ABTS-RSA) assay

ABTS-radical scavenging activity (ABTS-RSA) assay is based on the ability of an antioxidant to stabilize the blue-green colored ABTS+ cationic radical by transforming it into colorless ABTS by trapping a proton by the antioxidant. The decrease in absorbance caused by the antioxidant reflects the ability to capture the free radical. The decrease in absorbance (% inhibition) of the ABTS+ cationic radical solution translates the effect of the antioxidant sample **(Re *et al*., 1999).**

The ABTS-RSA assay was performed accordind to the protocol described by Re et al. **(Re *et al*., 1999)**. $ABTS^{+}$ radical cation was generated by reacting 7 mM ABTS in water and 2.45 mM potassium persulfate (1:1, v/v) and allowing the mixture to stand for 12–16 h in the dark before use. The $ABTS^{+}$ radical cation solution was then diluted with methanol to obtain an absorbance of 0.700 ± 0.02 at 734 nm. Peach kernel extracts (50 µl) were mixed with 950 µl of the ABTS solution and then incubated in the dark for 7 min at room temperature. Absorbance was measured at 734 nm against a blank. Trolox was used as standard for tarcing calibration curve (annex VI) and results were expressed as milligrams equivalents of Trolox per 100g of dry matter (mg ET/100 DM).

4.8.3 Ferric reducing power (FRP) assay

Reducing power is the ability of the antioxidants present in the extract to reduce the ferric iron of the ferricyanide complex Fe3+ to ferrous iron Fe2+. The increase in absorbency of the mixture containing the extract indicates an increase in reducing power **(Chew *et al,* 2009).**

The ferric reducing power (FRP) assay was determined according to the protocol of Oyaizu **(Oyaizu, 1986)**. Peach kernel extract (300 µl) were mixed with 750 µl of sodium phosphate buffer (0.2M, pH 6.6) and 750 µl of potassium ferricyanide (1%). After incubation at 50°C for 20 min in a water bath, 750 µl of trichloroacetic acid (10%) was added to the mixture. A fraction of the reaction mixture (500 µl) was mixed with distilled water (500 µl) and ferric chloride (100 µl, 0.1%) and the absorbance was measured at 700 nm. A calibration curve was made using ascorbic acid as standard (Annex III) and results were expressed as mg equivalents of ascorbic acid per 100g dry matter (mg EAA/100 DM).

4.8.4 Total antioxidant activity (TAA) assay

Total antioxidant activity (TAA) assay is based on the reduction of molybdenum Mo (VI) present in the form of molybdate ions MoO42- to molybdenum Mo (V) MoO2+ in the presence of the extract to form a green phosphate/Mo(V) complex at pH acid **(Prieto*et al.,* 1999).**

The total antioxidant activity (TAA) of the extracts is evaluated by the phosphomolybdenum method of **(Prieto *et al.,* 1999)**. Peach kernel extracts (100 µl) were mixed with 1 ml of the reagent solution (0.6 M sulfuric acid, 28 mMsodium phosphate and 4 mM ammonium molybdate). The test tubes were incubated for 90 min under 95°C in a water bath. After cooling, the absorbance of the solutions is measured at 695 nm. The TAA was determined by referring to the calibration curve produced with ascorbic acid (annex IV) and results were expressed in mg equivalents of ascorbic acid per 100g of dry matter (EAA/100 g of DM).

5 Statistical analysis

All analyses were performed in four independent assays (n=4). The data were expressed as means ± standard deviations (SD). Statistical analysis was performed using the software STATISTICA 5.5 (StatSoft Inc., Oklahoma, USA). An analysis of variance (ANOVA) is applied completed by Tukey's test in order to determine the significant differences between the samples for each parameter. The degree of significance of the data is taken at the probability $p<0.05$. Pearson correlation analysis was performed to determine the correlation among variables.

Results and discussions

1 Solvent type extraction

The solubilization of compounds depends on their affinity with the solvent. The differences in polarity between the various solvents therefore make it possible to target specific groups of molecules. **(Royer *et al*., 2010)**

1.1 Total phenolic compound

In this study, diverse solvents (acetone, ethanol and methanol), in mixture with water, were used to extract phenolic compounds from peach kernels. Results showed that all the peach kernels extracts contained phenolic compounds and the content of these compounds varied according to each solvent used (Fig. 8A). Among the tested solvents, 60% acetone was significantly ($p < 0.05$) the most efficient for extracting TPC from peach kernels (149 mg GAE/100 g), followed by 60% ethanol and 60% methanol with TPC of 61,9 mg GAE/100 g and 52,7 mg GAE/100 g, respectively.

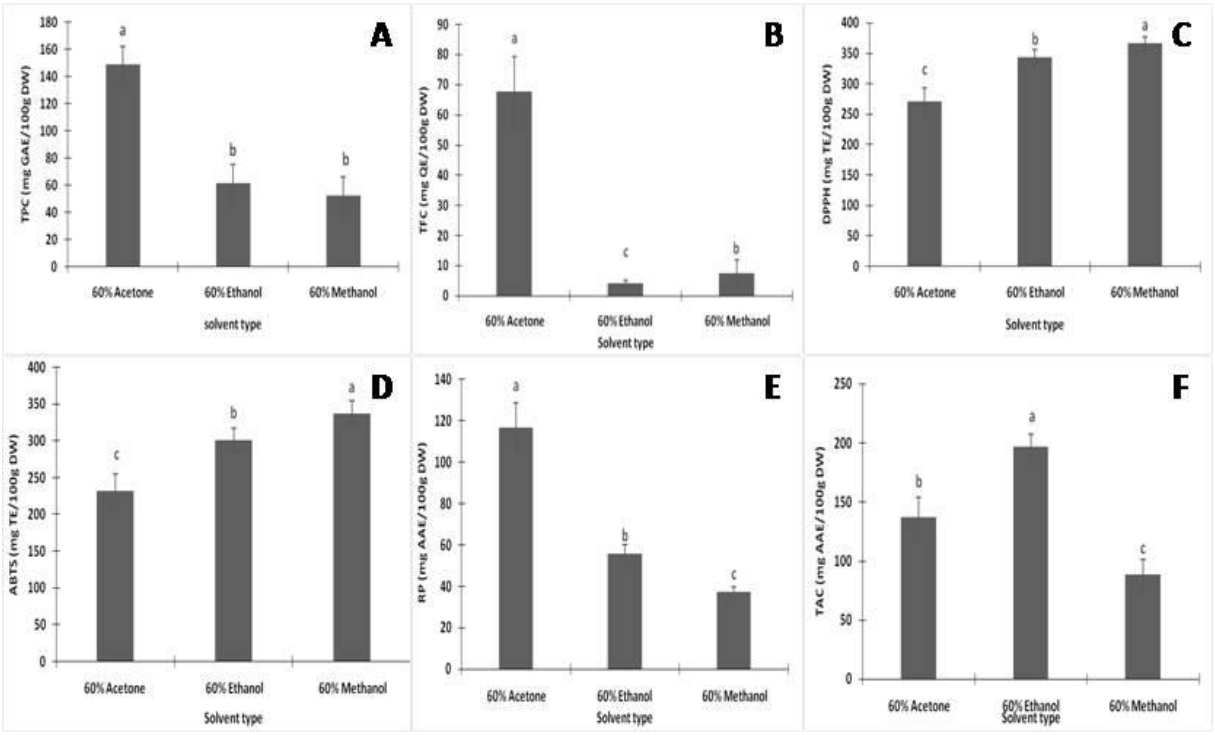


Figure 7:Effect of solvent type on the extraction of TPC (A), TFC (B), DPPH-RSA (C), ABTS-RSA (D), FRP (E) and TAC (F) from peach kernels.

The higher total phenolic content in 60% acetone solvent extraction may be due to the fact that major phenolic compounds of peach kernels are made of a long non-polar chain of carbon–carbon covalent bonds with a phenolic group attached to the two ends **(Shen *et al.*, 2012)**. The structure allows them to dissolve most freely in acetone with lower polarity, followed by ethanol and methanol.

The high efficiency of acetone to extract TPC from samples may be due to its ability to prevent the protein–polyphenol binding, which is insoluble complex, through solvent extraction **(Jakobek, 2015)**. It has been postulated that acetone is able to inhibit the formation of the protein–polyphenol complex during extraction, or to break down the interaction between the functional group of polyphenols (–OH) and the carboxyl group of proteins **(Hwang *et al.*, 2014)**.

Several studies are in agreement with our results and also found that acetone was the best solvent for the extraction of total phenolic compounds **(Sun *et al.*, 2005; Shui *et al.*, 2006; Liu *et al.*, 2007; Chavan *et al.*, 2013).**

1.2 Total flavonoid compound (TFC)

The solubility of flavonoids is highly correlated with their chemical structures and the nature of the extraction solvent **(Chebil*et al.*, 2007).** Among the different solvents studied (Figure 8B), 60% acetone (47 mg QE/100 g), was the best solvent for extracting flavonoids from the peach kernels, followed by 60% methanol (7,4 mg QE/100 g) and 60% ethanol (4,1 mg QE/100 g) (figure 8B).

Our findings were in agreement with the study of Ferreira and Pinhoreporting that the flavonoid (Hesperetin) was more soluble in acetone than in methanol, followed by ethanol **(Ferreira *et al.*, 2012)**.

1.3 DPPH -radical-scavenging activity (DPPH-RSA)

DPPH is a stable organic free radical, generally used to evaluate antiradical properties of any substance in vitro **(Bozin*et al.*, 2008)**. In the presence of free radical scavengers, DPPH

(2,2 diphenyl-1-picrylhydrazyl) from purple color (oxidized form) is reduced to yellow-colored 2,2 Diphenyl-1-picryl hydrazine. The intensity of the coloration, measured with a spectrophotometer at 517 nanometers, is inversely proportional to the anti-radical activity of the compounds whose wishes to determine the activity **(Kouamé et al., 2009).**

Scavenging stable DPPH° free radical test revealed that 60% methanol was observed to be the solvent exhibiting the highest antioxidant activity (367 mg TE/100 g), followed by 60% ethanol (343,7 mg TE/100g) and 60% acetone (270,5mg TE/100g) (figure 8C).

Our results are in agreement with **several** studies showing that methanol was the solvent presenting the highest antioxidant activity for the DPPH-RSA test **(Fan *et al*., 1997, Guo *et al*., 2001, Wong *et al*., 1997)**. Like our study, they also found that acetone extracts showed the weakest radical scavenging activity.

1.4 ABTS radical-scavenging activity (ABTS-RSA)

This assay is based on the neutralization of a radical-cation resulting from the single electron and the oxidation of the synthetic chromophore (acid 2,2'- azino-bis 3 ethylbenzothiazoline-6-sulfonic). This reaction is monitored spectrophotometrically by the change in the absorption spectrum **(Pellegrini*et al*., 1999).**

ABTS radical-scavenging activity assay was chosen because it can be applied to determine antioxidant capacity of both hydrophilic and hydrophobic antioxidants of plant extracts **(Dai *et al*., 2010)**

ABTS-RSA also revealed that 60% methanol was observed to be the solvent presenting the highest antioxidant activity (337 mg TE/100 g), followed by 60% ethanol (307,5,7 mg TE/100g) and 60% acetone (232,4mg TE/100g) (figure 8D).

Our results are in accordance with the study of Robles-Ramírez et al.who found that the highest antioxidant activity was obtained from the extracts with 80% methanol and 80% ethanol, while the values obtained with 80% acetonewere very low **(Robles-Ramírez*et al*., 2016).**

1.5 Ferric reducing power (FRP)

This power measures the ability of an antioxidant to reduce ferric iron Fe^{3+} (FeCl3) to iron ferrous Fe^{2+} (FeCl2) in the presence of a chromogenic agent potassium ferricyanide K3 [Fe(CN)6]. This results in the change from the yellow color of potassium ferricyanide to a blue-green color whose intensity depends on the reducing power of the antioxidant **(Chou *et al.*, 2003).**

Unlike ABTS-RSA and DPPH-RSA, results showed that 60% acetone extract was observed to have significantly the highest RP (116,5mg AAE/100 g). 60% ethanol and 60% methanol showed no significant differences with intermediate values of 55,8 and 37,2 mg AAE/100g, respectively (figure 8E).

Our results are in accordance of those of Chaalal et al.**(Chaalal*et al.,* 2012)**and Benchikh and Louailèche **(Benchikh *et al,* 2014)**, who found that 75% acetone exhibited the highest FRP activity.

1.6 Total antioxidant activity (TAA)

TAA assay is utilized for the spectrofotometric quantitation of total antioxidant capacity and employs cost-effective reagents. It based on the reduction of Mo(VI) to Mo (V) in presence of antioxidant compounds and subsequent formation of a green phosphate/Mo (V) complex at acidic pH and at higher temperature **(Prieto*et al.*, 1999).**

TAA revealed that 60 % ethanol was observed to be the solvent presenting the highest antioxidant activity (196,9 mg AAE/100 g), followed by 60 % acetone (137,2 mg AAE/100g) and 60% methanol (88,3 mg AAE/100g) (figure 8F).

Similar to the present study,Leccese et al. reported that ethanol extract showed the highest TAA value in abricot fruit **(Lecceseet *al*., 2011).**

Differences observed in the antioxidant activity (DPPH, ABTS, RP and TCA) of peach kernels extracts could be due to that the antioxidant activity of a phenolic extract cannot be predicted only on the basis of its total phenolic content, but it can be influenced by specific phenolic compounds present in this mixture **(Kähkönen*et al*., 1999)**.

Since 60% acetone gave the highest TPC and TFC yields, it was selected as the best solvent for extracting phenolic compounds and antioxidant activities from peach kernels for the following steps.

2 Solvent concentration

Combinations of solvents such as methanol, ethanol and acetone with water improve the extraction of phenolic compounds. **(Telli*et al*., 2010)**

2.1 Total phenolic compounds

Peach kernel's phenolic compounds were extracted using different concentrations of acetone (optimum solvent previously determined) ranging from 40% to 80%. Results showed that all acetone concentrations were capable of extracting phenolics as shown in figure 9A. The highest yield of TPC was recorded for 60% acetone (149 mg GAE/100 g), followed by 80% acetone (145,5 mg GAE/100 g) and 40% acetone (145,3 mg GAE/100 g) with no significant differences. Thus, 60% acetone was chosen as the best acetone gradient for the following steps.

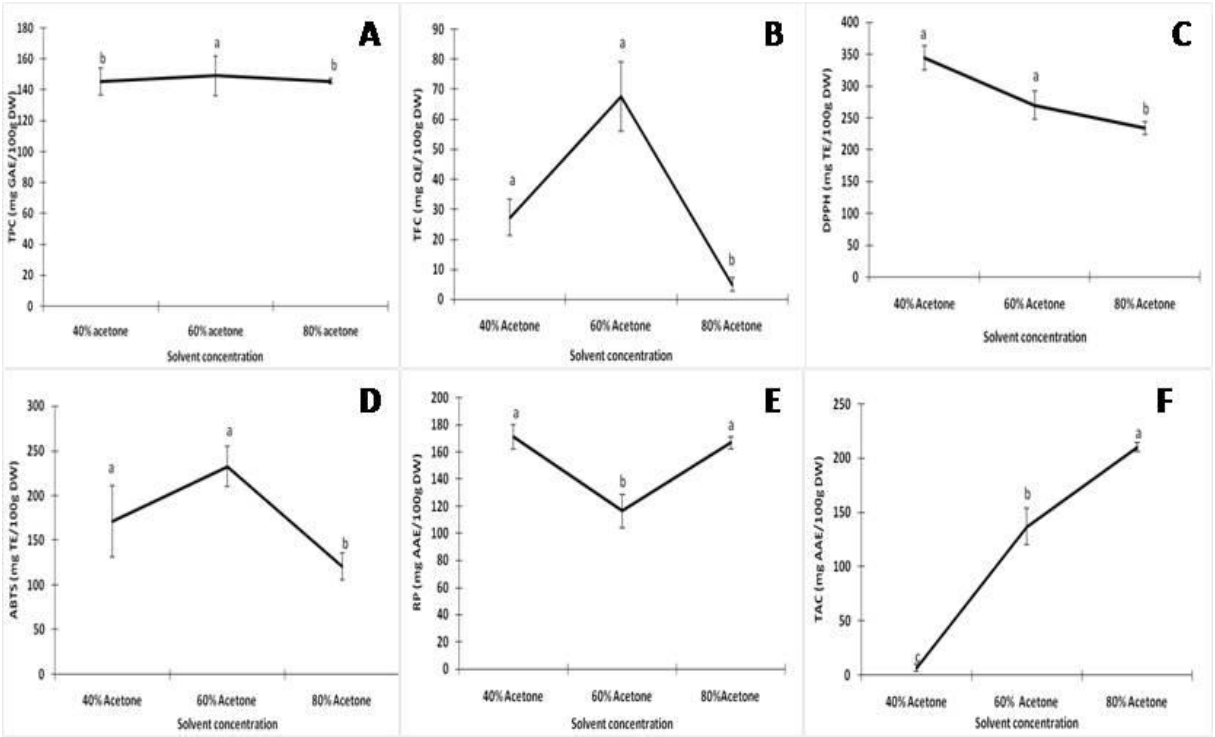


Figure 8:Effect of solvent concentration on the extraction of TPC (A), TFC (B), DPPH-RSA (C), ABTS-RSA (D), RP (E) and TAC (F) from peach kernels.

Our results are in accordance of those many previous studies. Uma et al. found that 60% is the best concentration of acetone for extracting TPC **(Uma*et al*., 2010).**Hobbi et al. demonstrated that 60% acetone was the best solvent for extracting antioxidant phenolic compounds from apple pomace**(Hobbi*et al*., 2021)**.

According to some authors, the addition of water to organic solvents such as acetone, methanol, and ethanol, creates a more polar medium that facilitates the extraction of phenolic compounds **(Spigno*et al.*, 2007)**. However, the use of pure water as solvent is not efficient to extract phenols because these compounds are often more soluble in organic solvents less polar than water **(Kim*et al.*, 2005).**

2.2 Total flavonoid compounds (TFC)

Results showed that all acetone concentrations were capable of extracting TFC with significant differences ($p < 0.05$) as shown in figure 9B. Same as TPC, the highest yield of TFC was recorded for 60% acetone (67,6 mg GAE/100 g), followed by 40% acetone (27,3 mg GAE/100 g) and 80% acetone (5 mg GAE/100 g).

Our findings were in agreement with the study conducted bySrivastava et al.in which 70% was found to be the best acetone concentration for extracting TFC from Feronia Limonia fruit **(Mishra *et al.*, 2020)**.

2.3 DPPH radical-scavenging activity (DPPH-RSA)

Results showed that acetone concentration had significant effect ($p < 0.05$) on DPPH-RSA of peach kernels extracts (Fig. 9C). The highest values were obtained at 40% acetone with (344,9 mg TE/100) followed by 60% acetone with (270,5 mg TE/100) and 80% acetone with (234,3 mg TE/100)

Our results are in accordance with the literature. Mashkor found that 50% was the best acetone concentration as it exhibited the highest DPPH-RSA in a study carried out on fenugreek seeds**(Mashkor, 2014**).Ikram et al showed that 40% acetone was the best acetone gradient with manifest the biggest DPPH-RSA **(Ikram*et al.*, 2020).**

2.4 ABTS radical-scavenging activity (ABTS-RSA)

Results of the ABTS-RSA test revealed that 60% acetone presented the highest of ABTS-RSA value (232,4 mg TE/100g) followed by 40% acetone (171,4 mg TE/100) 80% acetone with (121,1 mg TE/100). Note that no significant difference was observed 60% acetone and 40% acetone ABTS-RSA values (figure 9D).

Our findings were in agreement with the study of Jun et al. who found that the 60% acetone was the best acetone concentration for ABTS-RSA **(Jun*et al.*, 2014).**

2.5 Reducing power:

As showed in figure 9E, 40% acetone extract was observed to have significantly the highest RP (171,5mg AAE/100 g), followed by 80% acetone (166,7mg AAE/100 g). While 60% acetone exhibited the weak RP value (116,5mg AAE/100 g).

Similar to present study**,** Singh et al.**(Singh*et al.*, 2016)** reported that 50% acetone was the best acetone concentration manifesting the highest RP value.

2.6 Total antioxidant activity (TAA)

Total antioxidant activity revealed that 80 % acetone was observed to be the solvent presenting the highest antioxidant activity (210,2 mg AAE/100 g), followed by 60% acetone (137,2 mg AAE/100g) and 40% acetone with (6,9 mg AAE/100g).

(Kankara *et al,* 2016) study was in agreement with our results and demonstrated that 75% acetone extract has the highest value of total antioxidant activity (TAC).

3 Extraction time

The duration is closely linked to the extraction kinetics. Knowledge of the kinetics will make it possible to stop the extraction when the desired yield is reached and not to continue the operation. **(Mandal*et al.*, 2007)**

3.1 Total phenolic compounds (TPC)

Extraction process was carried out under different extraction times ranging from 3 to 60 min, as shown in Fig 10A. When the extraction time varied from 3 to 5 min, the variation on the extraction rate was relatively unchanged. Then, from 5 to 30 min, the TPC gradually increase, until reaching a peak at 30 min with a rate of (149 mg GAE/100 g). After extraction time exceeded 30 min, the variation on the TPC extraction rate decreased significantly. This may be due to the chemical reactions induced on the longer extraction time and which caused oxidative conversion of polyphenols.

Reasonably, a long extraction time causes the enhancement of extraction efficiency. However, longer extraction time increases the chance of oxidation of phenolics unless reducing agents are added to the solvent system **(Naczk *et al.* , 2004).**

Nevertheless, the time required to extract the active compounds depends on the amount of active compounds to be extracted and the solvent used. The higher the content of active compounds, the longer the extraction time required. **(Laily*et al.*, 2015)**

Our results are in accordance with those of Altemimi et al.**(Altemimi*et al.*, 2016)** who demonstrated that 25.67 min and 27.86 min was the best time for extracting phenolics from pumpkins and peach fruits, respectively.

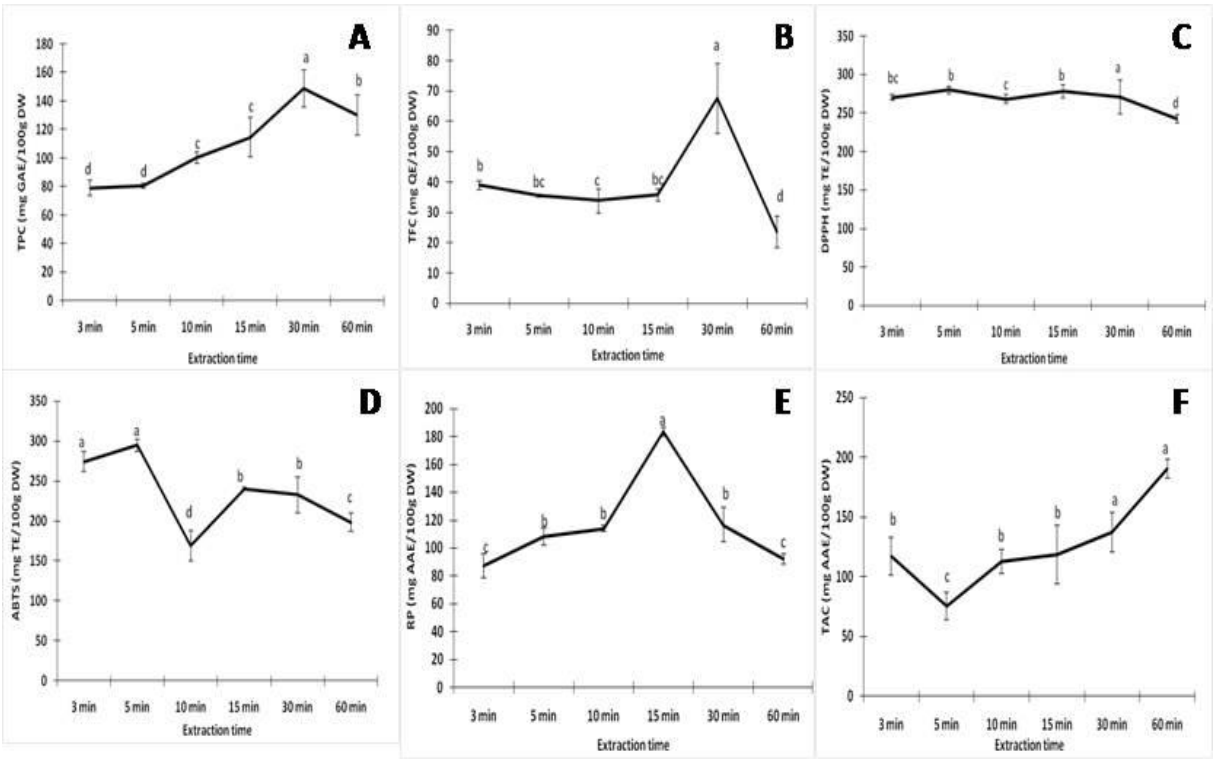


Figure9:Effect of the variation of the time on the extraction of TPC (A), TFC (B), DPPH-RSA (C), ABTS-RSA (D), RP (E) and TAC (F) from peach kernels.

3.2 Total flavonoid compounds (TFC)

The effect of extraction time (3.5, 10, 15, 30, 60 min) on the extraction yield of total flavonoids compounds (TFC) was presented in Fig 10B. Results showed that the yield of TFC was increased with the increasing of extraction time from 3 min to 30 min. Over 30 min, the yield significantly decreased. This might be due to the decomposition of active compounds during the prolonged extraction time **(Li,*et al.*, 2009; Sun *et al.*, 2010 Sheng *et al.*, 2011).**

Our findings were in agreement with the study conducted by Liu et al. in which 30 min was found the be the best extraction time for extracting TFC from *Gynura medica* leaf **(Liu*et al.*, 2010).**

3.3 DPPH radical-scavenging activity (DPPH-RSA)

Simillary to the TFC and TPC, the yield of scavenging stable DPPH° free radical was increased with the increasing of time extraction until reaching a maximum at 30 min (67,6 mg ET/100 g), over 30 min the yield slightly decreased.

Our findings were in agreement with the study ofAltemimiet al. which found that 30 min was the extraction time presenting the highest DPPH-RSA in spinach extract**(Altemimi*et al*., 2015).**

3.4 ABTS radical-scavenging activity

The yield of scavenging ABTS°+ free radical increased with the increasing extraction time until reaching a maximum at 5 min (279.7 mg TE/100 g). After extraction time exceeded 5 min, the variation ABTA-RSA rate decreased significantly and reached the lowest value at 10 min, then the variation on the ABTS-RSA remains unchanged when the extraction time ranged from 10 to 60 min (figure 10D).

Our results were in agreement with the study of Bhuyan et al.which found that the time ranging from 2 to 6 min was the best extraction time exhibiting the highest ABTS-RSA from Eucalyptus robusta **(Bhuyan*et al*., 2015).**

3.5 Reducing power

As shown in Fig 10E, reducing power's yield kept increasing from 3 to 15 min until reaching a maximum at 15 min (183,4 mg AAE/100 g). From 15 to 60 min, the variation on the extraction rate decreased significantly.

Our findings were in agreement with a the study ofWong et al. which found that 15 min was the extraction time exibiting the highest reducing power in kenaf seeds**(Wong*et al*., 2014).**

3.6 Total antioxidant activity (TAA)

Regarding TAA, as shown in Fig 10F, the variation on this activity continued to increase as the extraction time increases until reaching a maximum value at 60 min (190.5 mg AAE/100 g).

Our findings were in agreement with a the study ofLiyana-Pathirana and Shahidi**(Liyana-Pathirana *et al* 2005)** which demonstrated that 60 min extraction time manifested the highest TAA value.

Since the extraction time (30 min) gave the highest TPC yield, it was selected as the best extraction time for extracting phenolic compounds and antioxidant activities from peach kernels.

4 Extraction temperature

The rise in temperature allows the increase of the solubility and the diffusivity of the solute and the decrease of the viscosity. It must be limited to avoid the risk of extraction of harmful compounds and thermal degradation of the solute. **(Leybros *et al,* 1990).**

4.1 Total phenolic compounds

To study the effect of different temperatures on the extraction of polyphenols from peach kernels, the extraction process was carried out under different extraction temperature ranging from from 25 to 60°C, while other extraction condition was as follows: a ratio of solid/liquid (750 mg/ 30mL), acetone concentrations 60%, extraction time 30 min.

Results Fig 11A showed that increasing temperature from 25°C to 35°C, the TPC yield decreased slightly with not significant differences. For temperature varying from 35°C to 60°C, TPC increased gradually until reaching a maximum at 60°C (185,7mg GAE/100 g). Increased temperature promotes solvent extraction by enhancing both diffusion coefficients and the solubility of polyphenol content **(Al-Farsi *et al,* 2008).** Increased solubility of polyphenol contents was also reported by Wang et al. who found that increasing temperature favored the release of bound polyphenol in a sample with the breakdown of cellular constituents of plant cells which leads to increased cell membrane permeability**(Wang*et al.,* 2008).** Moreover, it is also believed that release of these bound polyphenols could further reduce the chances of polyphenols coagulating with lipoprotein, thereby enhancing solubility of the polyphenols and diffusion increasing polyphenol yield **(Zhang *et al*., 2007; Al-Farsi *et al,* 2008).**

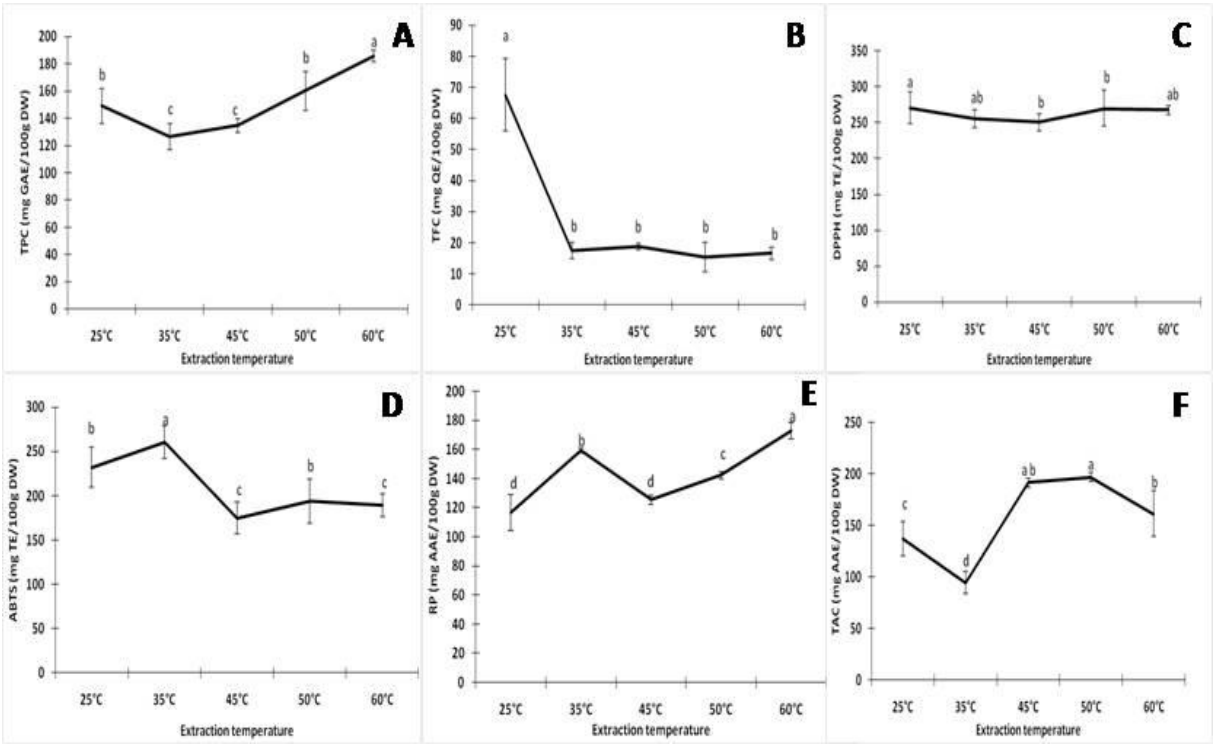


Figure10:Effect of the variation of the temperature on the extraction of TPC (A), TFC (B), DPPH-RSA (C), ABTS-RSA (D), RP (E) and TAC (F) from peach kernels.

4.2 Total flavonoid compounds

Fig 11B indicated that the yield of total flavonoids decrease with the increasing temperature, from 25°C which is the optimum temperature for TFC extraction (67,6 mg GAE/100 g) to 35°C. However, varying temperatures from 35°C to 60°C dosen't have any significant effect on TFC extraction. This could be explained by the degradation of some thermo-sensitive flavonoides compounds under high temperatures as reported byPrommuak et al.**(Prommuak*et al*., 2008)**and Trabelsi et al.**(Trabelsi*et al*., 2010).**

Our findings were in agreement with the study of Ismail et al.who found that 25°C was the best extraction temperature for extracting TFC from baobab fruit pulp **(Ismail *et al*., 2020).**

4.3 DPPH radical-scavenging activity

The DPPH-RSA decreased slightly when increasing the extraction temperature from 25°C to 60°C with a maximum value at 25°C (270.5 mg ET/100 g).

Our results were in agreement with the study ofQu et al.who found that 25°C was the extraction temperature with the highest DPPH-RSA activity from fermented black tea **(Qu *et al.*, 2020).**

4.4 ABTS radical-scavenging activity

Fig 11D showed that the ABTS-RSA increased gradually until reaching a maximum at 35°C, then it decreased from 35 to 45°C. After 45°C, the variation on the ABTS-RSA was unchanged.

Our results were in agreement with the study ofYim et al.who found that 35°C was the extraction temperature which presented the highest ABTS-RSA from Schizophyllum commune aqueous extract **(Yim *et al*., 2013).**

4.5 Reducing power

Fig 11E showed that the reducing power increased with the increasing extraction temperature until reaching 35°C. From 35°C to 45°C, reducing power decreased slightly, and then it continues to increase gradually until reaching a maximum at 60°C (173 mg AAE/100 g).

Our findings were in agreement with the study of Irakli et al. who found that 60°C was the extraction temperature which presented the highest reducing power in olive leaves**(Irakli*et al*., 2018).**

4.6 Total antioxidant activity (TAA)

The TAA decreased from 25°C to 35°C. Then it increased gradually until reaching a maximum at 50°C (196.5 mg AAE/100 g). After 50°C, the TAA decreased slightly at 60°C with no significant differences.

Our results were in agreement with the study of Maran et al.**(Maran *et al.*, 2017)** who demonstrated that 50°C extraction temperature manifested the highest TAA value.

Losses in antioxidant capacity of plant samples are often reported following a thermal treatment, likely due to the degradation of polyphenols which were previously mobilised at lower temperature. It is interesting to note that in the present study, increasing extraction temperature had a positive effect to both RP and TCA capacities. From this scenario, it is believed that the phenolic compounds present in Prunus persica kernels are thermally stable and that the extraction time selected in the previous stage is suitable for both moderate to high temperature without leading to unfavourable degradation **(Chan*et al*., 2009, Liyana-Pathirana *et al*., 2005).**

Since elevated temperature may not be suitable for all kinds of phenolic compounds and taking in consideration the aspect of energy consumption and cost at the industrial level, the temperature of 25°C was selected as the best extraction temperature.

5 The solid/liquid ratio

It is necessary to define the optimal ratio between the quantity of dry matter to be extracted and the volume of the solvent. The effect of this parameter is related to the principles of mass transfer, therefore the concentration gradient between the solid and the liquid. **(Pinelo*et al.*, 2005)**

5.1 Total phenolic compounds

To study the different solid –liquid ratio on the extraction yield of polyphenols, the extraction process was carried out using different ratio of solid/liquid: (50mg/30ml), (100mg/30ml), (250mg/30ml), (500mg/30ml) and (750mg/30ml), while other extraction conditions was as follows: acetone concentrations 60%, extraction time 30 min, the temperature 25°C by ultrasonication.

As shown in Fig 12A, increasing solid/liquid induced increasing in the TPC extraction yield until reaching a maximum at 750 mg/30 ml (149 mg GAE/100 g).

Our results were in agreement with the study ofJiang et al.reported that rising solid–liquid ratio induced increasing in the polyphenols tiled extraction **(Jiang*et al.*, 2007)**.

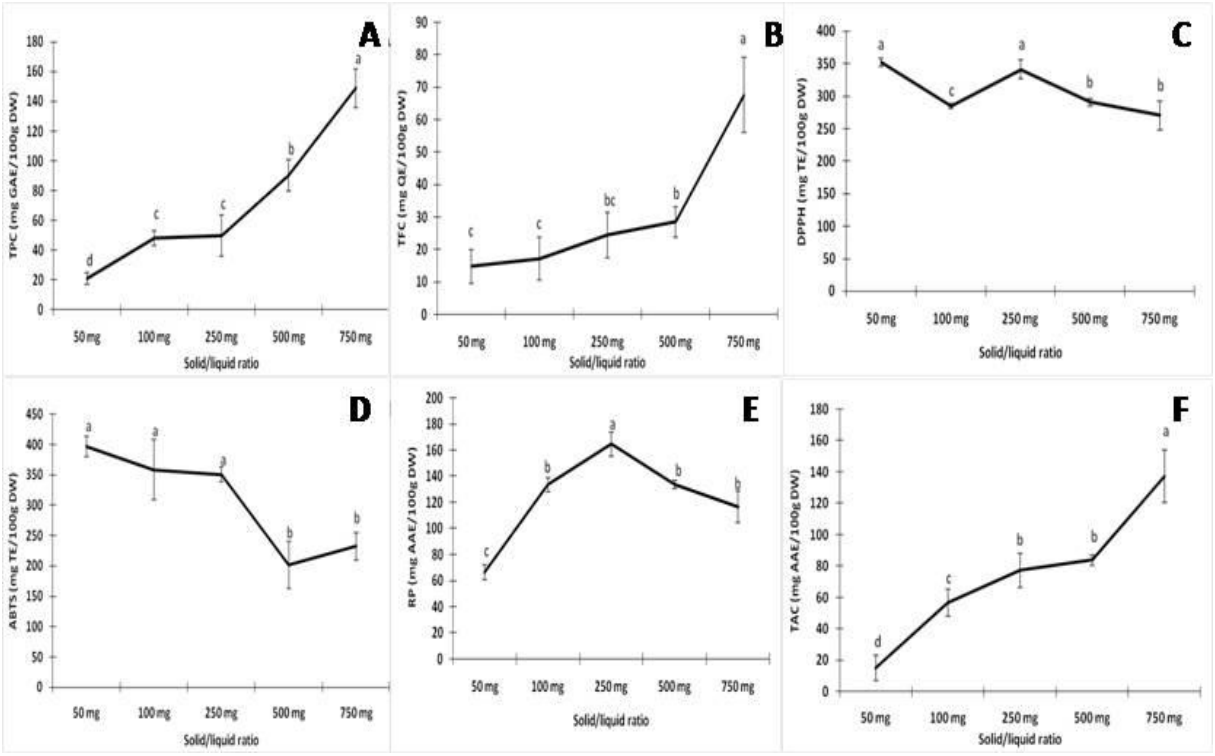


Figure11:Effect of solid/liquid ratio on the extraction of TPC (A), TFC (B), DPPH-RSA (C), ABTS-RSA (D), RP (E) and TAC (F) from peach kernels.

5.2 Total flavonoid compounds :

Similarly to polyphenols, the fig 12B indicated that the TFC extraction yield increased when the solid/liquid ratio increased until reaching a maximum at 750mg/30 ml ratio (67,6 mg QE/100 g).

5.3 DPPH radical-scavenging activity

Fig 12C showed that the best DPPH-RSA was registered at the ratio 50mg/30ml (352.3 mg ET/100 g), after which the DPPH-RSA decreased to 100mg/30ml ratio. Then it increasesd gradually until reaching the ratio of 250mg/30ml. After that, the DPPH-RSA deacreased slightly until reaching the ratio of 750mg/ml.

5.4 ABTS radical-scavenging activity

Similarly to DPPH-RSA, ABTS-RSA exhibited the highest value at the ratio of 50mg/30ml (396.9 mg ET/100 g), then it decreased until reaching 500mg/30ml ratio where it has the minimal rate. After that, the ABTS-RSA increased slightly to 750mg/30ml ratio (figure 12D).

5.5 Reducing power:

Results showed that reducing power increased with the increased of extraction ratio until reaching 250mg/30ml where it has the optimal value (164.5 mg AAE/100 g). Then it continues to decrease gradually until reachingthe ratio of 750 mg/30ml (figure 12E).

5.6 Total antioxidant activity (TAA)

For the TAA, the fig 12F indicated that the TAA continued to increase as the solid/liquid ratio increased until reaching its maximum value at the ratio of 750mg/30 ml (137,2 mg AAE/100 g).

6 Pearson correlation analysis:

6.1 Solvent type:

Tableau V:Pearson correlations coefficient between different assays under influence of solvent type.

	TFC	DPPH	ABTS	RP	TAC
TPC	0. 89***	-0. 74**	-0.83***	0. 89***	0.13^{ns}
TFC		-0.89***	-0. 85***	0. 94***	-0.20^{ns}
DPPH			0.84***	-0. 94***	0.07^{ns}
ABTS				-0.86***	-0.11^{ns}
RP					0.06^{ns}

ns Not significant.
* Significant at $p < 0.05$.
** Significant at $p < 0.01$.
*** Significant at $p < 0.001$

TPC, and DPPH-RSA (r=-0.74) and ABTS-RSA (r=-0.83). For TFC, good positive correlation (r=0, 94) was found between TFC and RP. But negative correlations were noted between TFC, and DPPH-RSA (r=-0.89) and ABTS-RSA (r=-0.85). These suggest that peach kenels phenolic compounds may act as reducing agents more than antiradical substances. No significant correlations were found between TPC, TFC and TCA assay.

It could be concluded that there are other phenolic compounds other than flavonoids and polyphénols witch contribute to the DPPH-RSA and ABTS-RSA of peach kernels extracts. However, correlations between TPC, TFC and RP antioxidant assay were positively high (r = 0,89 and r = 0,94 respectively at $P < 0.001$), we deduce that TPC and TFC may responsible for reducing power capacity.

6.2 Solvent concentration:

Tableau VI:Pearson correlations coefficient between different assays under influence of solvent concentration.

	TFC	DPPH	ABTS	RP	TAC
TPC	-0.04^{ns}	-0.01^{ns}	-0.53^{ns}	0. 74**	0.09^{ns}
TFC		0.56^{ns}	0.28^{ns}	-0.44^{ns}	-0.43^{ns}
DPPH			0.03^{ns}	0.14^{ns}	-0. 86***
ABTS				- 0.70**	-0.23^{ns}
RP					-0.01^{ns}

ns Not significant.
* Significant at $p < 0.05$.
** Significant at $p < 0.01$.
*** Significant at $p < 0.001$

Concerning the influence of solvent concentration (**Table VI**), only TPC was observed to be correlated positively with RP assay (r = 0.74, $P < 0.01$). No significant correlations were

found between the other assays. From this correlation, we can believe that peach kernels polyphenols are good reducing agents.

6.2.1 Extraction time:

Tableau VII:Pearson correlations coefficient between different assays under influence of extraction time.

	TFC	DPPH	ABTS	RP	TAC
TPC	0. 40 ^{ns}	-0.47*	-0. 43 ^{ns}	0. 05 ^{ns}	0. 71***
TFC		0.37 ^{ns}	0.13 ^{ns}	0.05 ^{ns}	-0. 17 ^{ns}
DPPH			0. 49*	0. 49*	-0.73***
ABTS				0. 17 ^{ns}	-0.46 ^{ns}
RP					-0. 16 ^{ns}

ns Not significant.
* Significant at p < 0.05.
** Significant at p < 0.01.
*** Significant at p < 0.001

Under the influence of extraction time (**Table VII**), TPC was found to be negatively correlated with DPPH-RSA assay (r = -0.47, P < 0.05). At the same time, a strong positive correlation was found between TPC and TAA assay (r = 0.71, P < 0.001), from these experimental results, we can believe that phenolic compounds are the contributors to the overall total antioxidant activity of peach kernels.

6.3 Extraction temperature:

Tableau VIII: Pearson correlations coefficient between different assays under influence of extraction temperature.

	TFC	DPPH	ABTS	RP	TAC
TPC	-0. 27 ^{ns}	-0.10 ^{ns}	-0.54*	0.73***	0.61**
TFC		0.10 ^{ns}	0.30 ^{ns}	-0.66**	-0.29 ^{ns}
DPPH			0.37 ^{ns}	0.01 ^{ns}	0.31
ABTS				-0.28 ^{ns}	-0.73***
RP					0.22 ^{ns}

ns Not significant.
* Significant at p < 0.05.
** Significant at p < 0.01.
*** Significant at p < 0.001

Under the influence of extraction temperature (**Table VIII**), TPC was correlated negatively with ABTS-RSA assay (r = -0.54) at P < 0.05 as well as TFC with RP assay (r = -0.66 , P < 0.01). However, strong correlations were found between TPC and RP and TAC assays, with Pearson correlations coefficient of r = 0.73 and r = 0.61 respectively at P < 0.01.

Therefore, we can believe that high temperatures did not affect the reducing capacity of peach kernels phenolics. Nevertheless, it might reduce their radical scavenging activity.

6.4 Solid/liquid ratio:

Tableau IXPearson correlations coefficient between different assays under influence of solid-liquid/ratio.

	TFC	DPPH	ABTS	RP	TAC
TPC	0.90***	-0.75***	-0.75***	0.21^{ns}	0.93***
TFC		-0.62**	-0.59**	0.15^{ns}	0.83***
DPPH			0.53*	-0.36^{ns}	-0.71**
ABTS				-0.08^{ns}	-0.66**
RP					0.45^{ns}

ns Not significant.
* Significant at $p < 0.05$.
** Significant at $p < 0.01$.
*** Significant at $p < 0.001$

Under the variation of solid/liquid ratio (**Table IX**), correlations between TPC, TFC and antioxidant activities (DPPH-RSA and ABTS-RSA) were negative with Pearson correlations coefficient of -0.75, -0.83 and -0.62, -0.59, respectively. It could be concluded that there are other antioxidant compounds other phenolics witch contribute to the DPPH-RSA and ABTS-RSA of peach kernels extracts. However, correlations between TPC, TFC and TCA antioxidant assay were positively high ($r = 0.93$ and $r = 0.83$, respectively at $P < 0.001$). No significant correlations were found between TPC, TFC and RP assay.

Conclusion and perspectives

Conclusion and perspectives

This study reports for the first time the optimization of the recovery of phenolic compounds from peach kernels by ultrasound-assisted extraction (UAE) technique using single factor experiments as experimental design. In total, five extractions parameters namely effect of solvent (60 acetone, 60% ethanol and 60% methanol), effect of acetone concentration (40%, 60% and 80%; v/v), extraction time (3, 5, 10, 15, 30 and 60 min), extraction temperature (25, 35, 45, 50 and 60°C) and solid/liquid ratio (50, 100, 250, 500 and 750 mg) were tested on the extraction of total phenolic compounds; TPC and total flavonoid compounds; TFC and on the antioxidant activities (DPPH-radical scavenging activity; DPPH- RSA, ABTS-radical scavenging activity; ABTS-RSA, reducing power; RP and total antioxidant activity; TAA).

Results showed that all extractions parameters tested affected significantly (P<0.05) the TPC, TFC and antioxidant activities (DPPH-RSA, ABTS-RSA, RP and TAA) of peach kernels. The optimal extractions parameters were sonication of 750 mg of sample with 30 ml of 60% acetone for 30 min at 25°C. These optimized conditions permitted an extraction yield of 149 mg GAE/100 g DM for TPC, 47 mg QE/100 g DM for TFC, 270,5mg TE/100g DM for DPPH-RSA, 232,4mg TE/100g DM for ABTS-RSA, 116,5mg AAE/100 g DM for RP and finally 137,2 mg AAE/100g DM for TAA.

Pearson correlation analysis showed, globally, good positive coefficients correlation between phenolic compounds and antioxidant activity (reducing power), particularly under the influence of solvent type and acetone concentration. These suggest that peach kernels phenolic compounds are good reducing agents. Strong positive correlation was found between TPC and TAA assay (r = 0.71, P < 0.001) under the influence of time extraction, suggesting that phenolic compounds are the contributors to the overall total antioxidant activity of peach kernels. For temperature extraction parameter, positively high correlation were found between TPC and RP and TAC assays which is suggest that high temperatures did not affect the reducing capacity of antioxidant activity of peach kernels phenolic compounds. Finally, we can conclude that peach kernels are a good source of phenolic compounds displaying strong antioxidant activities (antiradical scavenger activity and reducing power) and therefore, peach kernels extracts may be used as potential functional ingredients or additives in food industry, cosmetics and medicine.

However, in order to complete this work, it would be interesting to:

- Investigate other factors and extraction conditions that may influence the optimization of extraction of bioactive compounds such as: pH, sonication frequency, peach variety which could possibly influence the yield of the phenolic compounds.
- Use advanced analysis techniques (HPLC, LC/MS, etc.) to identify the phenolic compounds responsible of the antioxidant activity of peach kernels.
- Study other biological activities such as antibacterial and antifungal activities.
- Evaluate the cytotoxicity effect of peach kernels phenolic extracts.
- Finally, to carry out in vivo tests of the optimal extracts in order to determine their effects on animal health such anti-inflammatory activity, anti-diabetic activity,ect.

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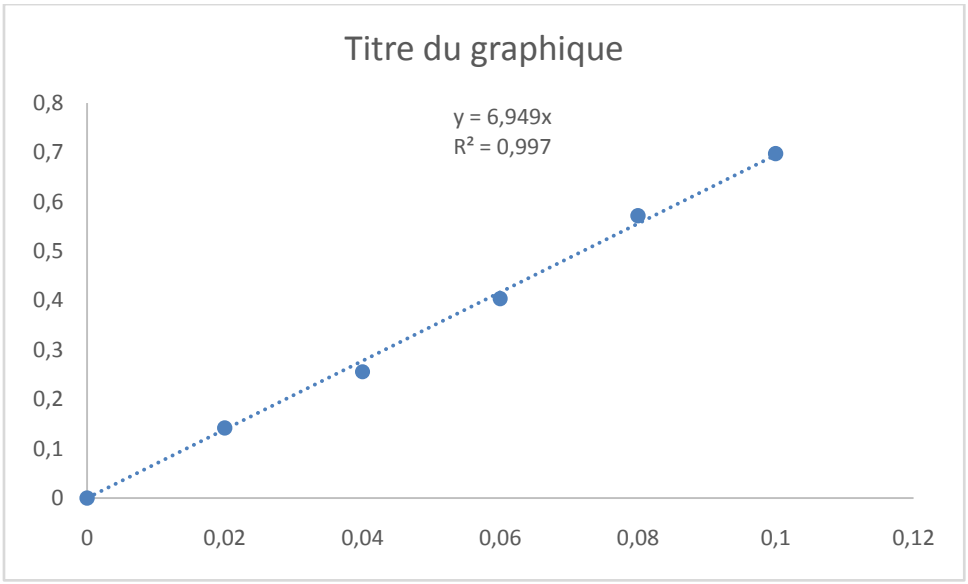
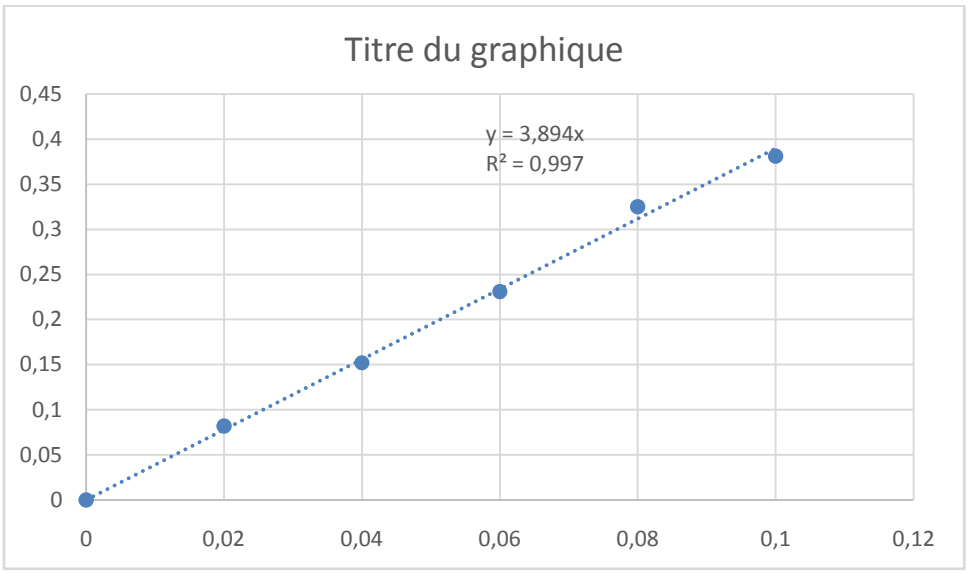
Annexes

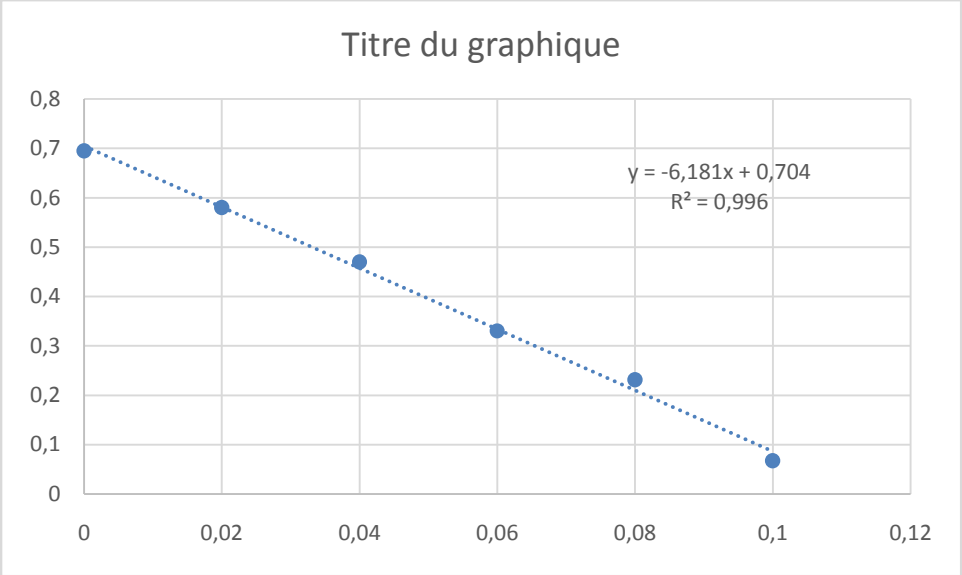
Annex I : Calibration curve of TPC.



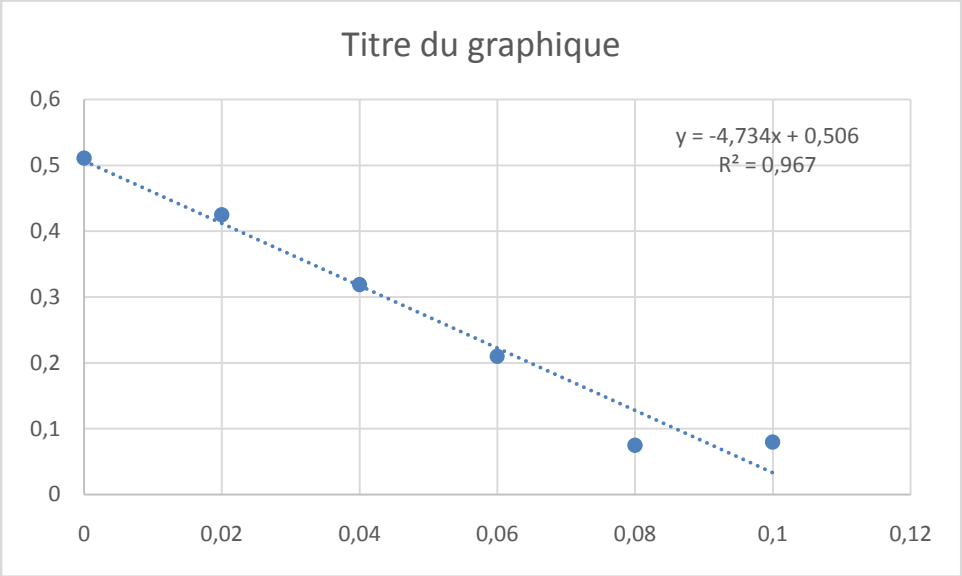
Annex II: Calibration curve of TFC.



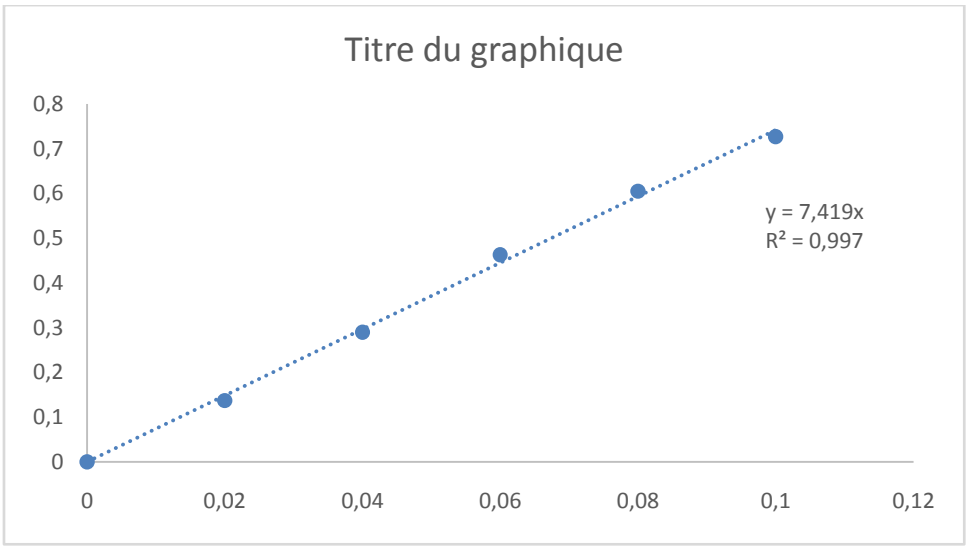
Annex III: Calibration curve of DPPH



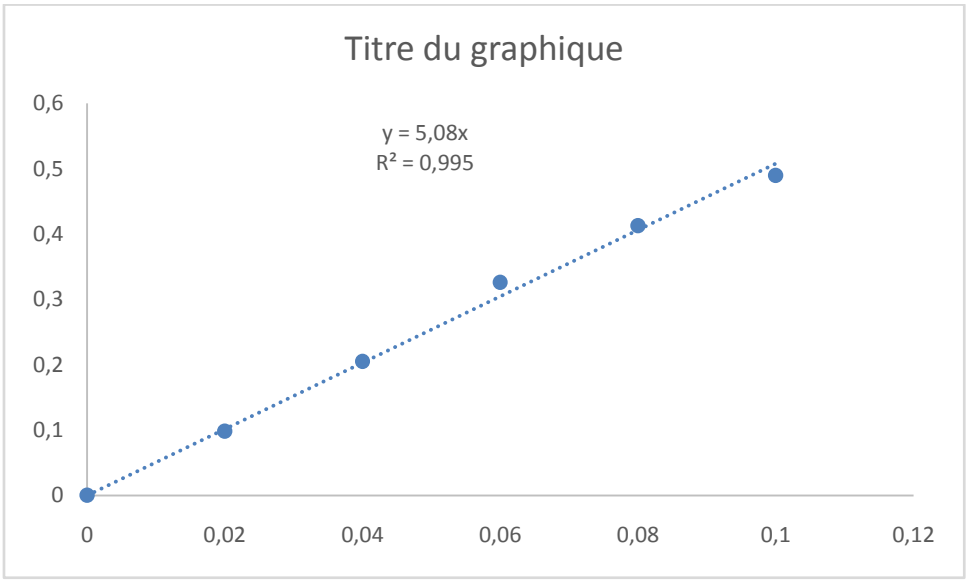
Annex IV: Calibration curve of ABTS



Annex V: Calibration curve of reducing power



Annex VI: Calibration curve of total antioxydant activity



Abstract:

This study reports the optimization of the recovery of phenolic compounds from peach kernels byultrasound-assisted extraction (UAE) technique using single factor experiments approach. Five extractions parameters namely effect of solvent (60 acetone, 60% ethanol and 60% methanol), effect of acetone concentration (40%, 60% and 80%; v/v), extraction time (3, 5, 10, 15, 30 and 60 min), extraction temperature (25, 35, 45, 50 and 60°C) and solid/liquid ratio (50, 100, 250, 500 and 750 mg) were tested on the extraction of total phenolic compounds; TPC and total flavonoid compounds; TFC and on the antioxidant activities (DPPH-radical scavenging activity; DPPH-RSA, ABTS-radical scavenging activity; ABTS-RSA, reducing power; RP and total antioxidant activity; TAA). Results showed that the optimal extractions parameters were sonication of 750 mg of sample with 30 ml of 60% acetone for 30 min at 25°C. These optimized conditions permitted an extraction yield of 149 mg GAE/100 g DM for TPC, 47 mg QE/100 g DM for TFC, 270,5mg TE/100g DM for DPPH-RSA, 232,4mg TE/100g DM for ABTS-RSA, 116,5mg AAE/100 g DM for RP and finally 137,2 mg AAE/100g DM for TAA. Pearson correlation analysis showed, globally, good positive coefficients correlation between phenolic compounds and antioxidant activities of peach kernels. It can be conclude that peach kernels are a good source of phenolic compounds displaying strong antioxidant activities (antiradical scavenger activity and reducing power) and therefore, peach kernels extracts may be used as potential functional ingredients or additives in food industry, cosmetics and medicine.

Keywords: peach kernels, phenolic compounds, antioxidant activity, optimization, extraction, sonication, solvent, time, temperature.

Résumé:

Cette étude rapporte l'optimisation de la récupération des composés phénoliques des noyaux de pêche par la technique d'extraction assistée par ultrasons (UAE) en utilisant une approche d'expériences à facteur unique. Cinq paramètres d'extractions à savoir effet du solvant (60 acétone, 60 % éthanol et 60 % méthanol), effet de la concentration en acétone (40 %, 60 % et 80 % ; v/v), temps d'extraction (3, 5, 10, 15, 30 et 60 min), la température d'extraction (25, 35, 45, 50 et 60°C) et le rapport solide/liquide (50, 100, 250, 500 et 750 mg) ont été testés sur l'extraction des composés phénoliques totaux ; CPT et composés flavonoïdes totaux ; CFT et sur les activités antioxydantes (activité de piégeage de radical DPPH; DPPH-RSA, activité de piégeage de radical ABTS ; ABTS-RSA, pouvoir réducteur ; RP et activité antioxydante totale ; AAT). Les résultats ont montré que les paramètres d'extraction optimaux étaient la sonication de 750 mg d'échantillon avec 30 ml d'acétone à 60 % pendant 30 min à 25 °C. Ces conditions optimisées ont permis un rendement d'extraction de 149 mg GAE/100 g DM pour le CPT, 47 mg QE/100 g DM pour les CFT, 270,5 mg TE/100 g DM pour le test DPPH-RSA, 232,4 mg TE/100 g DM pour le test ABTS- RSA, 116,5mg AAE/100g DM pour le RP et enfin 137,2mg AAE/100g DM pour l'AAT. L'analyse de corrélation de Pearson a montré, globalement, une bonne corrélation et des coefficients positifs entre les composés phénoliques et les activités antioxydantes des noyaux de pêche. On peut en conclure que les noyaux de pêche sont une bonne source de composés phénoliques présentant de fortes activités antioxydantes (activité antiradicalaire et pouvoir réducteur) et, par conséquent, les extraits de noyaux de pêche peuvent être utilisés comme ingrédients fonctionnels potentiels ou additifs dans l'industrie alimentaire, cosmétique et médicale.

Mots clés : noyaux de pêche, composés phénoliques, activité antioxydante, optimisation, extraction, sonication, solvant, temps, température.